KONINKLIJKE AKADEMIE VAN WETENSCHAPPEN
TE AMSTERDAM.

PROCEEDINGS

VOLUME XXV

Nos. 5 and 6.

President: Prof. F. A. F. C. WENT.
Secretary: Prof. L. BOLK.

(Translated from: "Verslag van de gewone vergaderingen der Wis- en Natuurkundige Afdeeling," Vol. XXXI).

CONTENTS.

H. ZWAARDEMAKER: "On the Alpha-automaticity of the Autonomous Organs", p. 152.

C. E. B. BREMEKAMP: "Further researches on the antiphototropic curvatures occurring in the coleoptiles of Avena". (Communicated by Prof. F. A. F. C. WENT), p. 158.

R. WEITZENBÖCK: "Ueber Wirkungsfunktionen". (Communicated by Prof. L. E. J. BROUWER), p. 166.

J. W. JANZEN and L. K. WOLFF: "Studies on the bacteriophagus of D'HERELLE". (Communicated by Prof. C. EYKMAN), p. 171.

A. W. K. DE JONG: "The Biscoumaric Acids", p. 175.

G. HERTZ: "On the Excitation and Ionization Potentials of Neon and Argon". (Communicated by Prof. P. EHRENFEST), p. 179.

C. A. H. VON WOLZOGEN KÜHR: "On the Occurrence of Sulphate-reduction in the deeper layers of the Earth". (Communicated by Prof. G. VAN ITERSON Jr.), p. 188.

E. VAN THIEL: "The Influence of a Catalyst on the Thermodynamic Quantities Regulating the Velocity of a Reaction". (Communicated by Prof. J. BÖESEKEN), p. 199.

J. BÖESEKEN: "The Dislocation Theory of Catalysis", p. 210.

W. E. DE MOL: "The disappearance of the diploid and triploid magnicoronate narcissi from the larger cultures and the appearance in their place of tetraploid forms". (Communicated by Prof. G. VAN ITERSON Jr.), p. 216.

L. J. SMID Jr.: "Numbers of Circles Touching Plane Curves Defined by Representation on Point Space". (Communicated by Prof. HENDRIK DE VRIES), p. 221.

A. F. HOLLEMAN: "Monochloro-trinitrobenzenes", p. 223.

A. A. WEINBERG: "On Respiratory Oscillations in the Galvanogram of Man". (Communicated by Prof. E. D. WIERSMA), p. 225.

Physiology. — *"On the Alpha-automaticity of the Autonomous Organs."* By Prof. H. ZWAARDEMAKER.

(Communicated at the meeting of June 24, 1922).

In the organism there are some organs which perform automatic movements and whose movements are continued also in parts that have been isolated from the body. Without any outward stimulation, simply by watching those parts we can follow up the continuation of this action in its causal and conditional relations. The type of such an organ is the heart. It is the musclecells themselves that pulsate, from the earliest embryonal existence up to death. Such a pulsating heart-cell is comparatively a simple system of phases, [1]) which, if the nucleus is left out of consideration, is made up of the following components: 1st. 7 ions, H, OH, Na, K, Ca, HCO_3, H_2PO_4 (resp. HPO_4); 2nd. 2 lipoids, cholesterin and lecithin; 3rd. a carbohydrate, glycogen, which is alternately combined with phosphoric acid and isolated from it again; 4th. oxygen; 5th. proteins and water as a solvent. The absolute quantity of every component exerts, according to the rules of the equilibrium of the phases, an influence upon the whole. Influence may be exerted, a component may even be given a certain concentration, by surrounding the cell with a nutrient liquid composed for the purpose. In so doing substitution appeared to be possible. Na may be replaced by Li or by highly purified Cs; K by all radio-active elements [2]); Ca by Sr and Ba; lecithin by sodiumoleonate. Besides the absolute quantity also the mutual relations carry weight, notably H : OH, H : HCO_3, K : Ca. Such interrelations must keep within certain bounds. To test this various qualities of the function may be considered: first of all the so-called tonus-condition, i.e. the degree of continued contraction between the limits of atony and maximal tonus; next the excitability in the several intervals of a period; lastly the automatic movement itself. Now granting the conditions of the system to be so regulated that the bounds we alluded to, have been kept in view, and each

---

[1]) H. ZWAARDEMAKER, Erg. des Physiol. Bd. 5. p. 135. 1906.

[2]) H. ZWAARDEMAKER, These Proceedings Vol. XIX, p. 633. (1916).

of the three fundamental manifestations, i.e. tonus, excitability and automaticity persist freely, two sorts of automaticity can be elicited by superadding successively the several radioactive elements to the nutrient liquid which surrounds the cells. Two sorts we say, because there are two groups of radio-active elements, which thus far I have been able to use as medium in the solutions of RINGER or TYRODE to substitute vice versa: 1st. an α-group: uranium, radium, emanation, polonium, thorium, 2nd a β-group: potassium, rubidium.

We shall now discuss the points of distinction and of agreement between these alpha-, and beta-automaticities.

The principal feature of an automatic, periodic movement is its tempo, which in its turn depends again on the so-called refractory stage inserted into every period. Now this tempo is determined by the amount of radio-activity for the alpha-group, as well as for the beta-group. A minimum amount is required for the movements to reveal themselves at all, and a maximum quantum that should on no account be surpassed. This allows a certain latitute for dosages, which is narrow for the alpha-, and broad for the beta-group.

Somewhere in this latitude there is a point of greatest frequency, the optimum. This point being established for the two sorts of rayers, the frequencies will be the same for either group.

Such an investigation evidently requires a constant temperature. It is also clear that, when the temperature is variable the two determining factors: amount of radio-active matter and degree of the temperature, may cooperate or counteract each other. It has already been shown that there is a law, which determines these relations, but I do not intend to enter into it here. Now, when both for potassium and for uranium the optimum doses have been found, which yield the highest frequency, the frequencies for potassium- and for uranium-automaticity are equal. This is instanced in Fig. 1. In the centre the potassium-beat is shown, separated to the right and to the left by a standstill from two other pulsations; these two other pulsations represent the paradoxical phenomenon appearing when passing from perfect potassium-dosis to a perfect uranium-dosis, and conversely [1]). To the right and to the left the uranium-beat can be observed. Its frequency does not differ from that of the automaticity in the centre of the figure.

Another property the two automaticities have in common is a

[1]) These Proceedings Vol. XIX, p. 1043, (1917) C.R. Soc. de Biol. t. 84 p. 704. Paris 1921.

similar need for radio-activity in the several subdivisions of the heart. This is seen best when passing from a perfect uranium-dosis

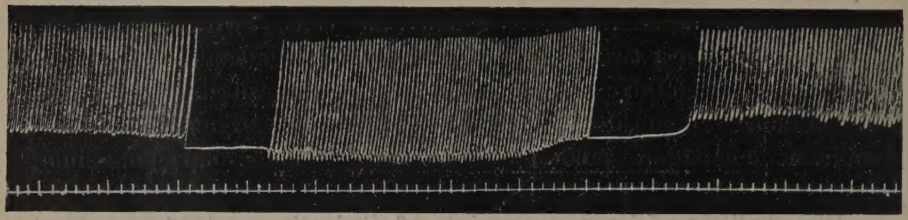


Fig. 1.

Frog's heart KRONECKER's canula 14° C., red light.

Transition from a circulating fluid with 25 mgr. uranylnitrate to another with 300 mgr. potassiumchlorid per litre, and then back again to 25 mgr. uranylnitrate. The transitions generally took place resp. 40 and 60 seconds before the paradoxical standstills, indicated in the figure by a white line. Potassium pulsation in the centre. Time $^1/_6$ min.

to a perfect potassium-dosis or the reverse with a simultaneous registration of the sinus, atrium, ventricle. If no conductive disturbances occur, it will be seen that the three divisions of the heart will stand still and resume their beats at the same moment with automaticities of their own. This is illustrated below in fig. 2 for an eel-heart in situ, which was perfused first with a uranium-liquid and at a moment designated in the time-line by *S* with a potassium-liquid.

The phenomenon, instanced in fig. 2 requires, however, accurate dosage of uranium as well as of potassium. It would not be surprising, if inaccuracy in this respect should engender dromotropism.

A third property the two automaticities have in common is the self-regulation after extrasystole, for α-conditions as complete as for the β-conditions.

The fourth common property is the initial similarity of the alpha- and the beta-electrogram, though I must admit that afterwards a difference may come forth through secondary influences [1]).

Only in adventitious respects do the two automaticities differ.

Of the greatest importance in this respect is the tonicity of the heart. The conditions determining the auto-tonus of the cardiac muscle are:

---

1) Klinische Wochenschrift Jahrg. I N°. 12 (or Diss. H. SLOOFF, Utrecht 4 July 1922).

a. the number of calcium-ions placed or not placed over against univalent-ions.

b. the number of H-ions.

c. the amount of light incident upon the heart, especially in the presence of a fluorescent substance.

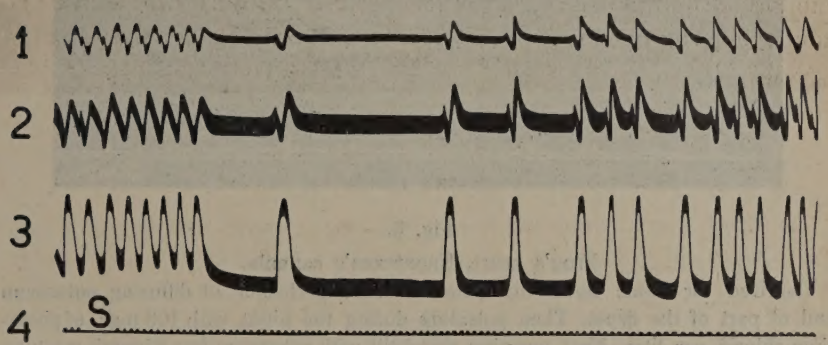


Fig. 2.

Eel's heart in situ. Perfusion from vena cava, first with a circulating fluid, containing 15 mgr. thorium-nitrate per litre, then with a circulating fluid, containing 100 mgr potassium-nitrate. 1 sinus, 2 atrium, 3 ventricle. Time in sec. At S transition from one fluid (thorium-beat) to the other (potassium-beat). Only in the ventricle a light tonus is noticeable during the thorium-beat. It has disappeared already for the greater part in the first beat performed by the heart during the paradoxon.

When applying uranium as an *a*-rayer each of the three above conditions is modified. Sub *a* undergoes a change because over against the calcium-ion not only univalent ions are placed, but also uranyl. Sub *b* is modified, because a solution of uranyl-salt causes a small increase of H-ions in Ringer's solution. True, this factor may be eliminated by the addition of a trace of $CaCO_3$, but let it be supposed that this did not take place. Sub *c* has been modified, because in the organ perfused with a potassium-fluid the incident light has only an inappreciable influence unless its strength be enormous, while in the presence of a fluorescent uranium-liquid also ordinary light will show its tonic action.

It will, therefore, be considered quite rational that in fig. 1 the bases of the uranium-elevations are not so low as those of the potassium-elevations. When thorium is substituted for uranium the phenomenon is less pronounced, still, it is certain that even then the tonus is not quite absent.

With emanation-beats [1]) and with pulsation evoked by outside radiations with polonium, there is often some increase of tonus,

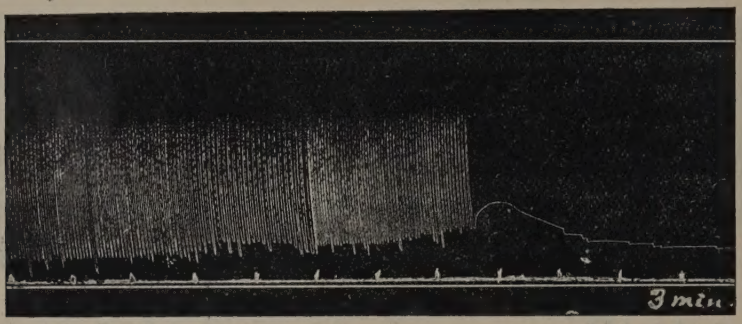


Fig. 3.

Frog's heart, KRONECKER's cannula.

Deprived for some hours by potassium-free perfusion of diffusing potassium and of part of the depot. Then pulsating during the night with 100 mgr. of potassium-chlorid per litre. Next morning standstill with potassium-free Ringer's mixture. Recovery of pulsation due to omnilateral polonium radiation. At the beginning of the curve the polonium was taken away.

Nearly half an hour later the polonium-beats cease. They had caused no increase of tonus worth mentioning.

which need not surprise us if we consider what has been stated sub *a*. Increase of tonus, however, is not a typical feature of alpha-automaticity, since it can exist without this increase when it is brought about from the outside by polonium-radiation. This is illustrated in Fig. 3 [2]). A heart that after cautious, prolonged perfusion with potassium-free Ringer's solution, had been deprived of a considerable portion of its potassium depot, continued pulsating for a long time also when subjected from the outside to omnilateral polonium-radiation. These pulsations occur without additional increase of tonus. The polonium is taken away at the beginning of the figure.

Besides in the tonus-condition, the two automaticities are also distinguished in the relation of the regularly pulsating hearts to the action of the constant current, to the alternating current and to diathermy. These distinctions have been described by Dr. DEN BOER [3]) in his Thesis, so that I will not revert to them.

---

[1]) ZWAARDEMAKER and T. P. FEENSTRA, C. R. Soc. de biologie, t. 84, p. 377. Paris 1921. ZWAARDEMAKER, Klin. Wochenschr. Jahrg. I, N°. 11. 1922. Arch. intern. de Physiol. vol. 18, p. 284, 1921.

[2]) Another instance is given by ZWAARDEMAKER and G. GRIJNS, Arch. néerland. de physiol. t. 2, p. 502, 1918.

[3]) M. DEN BOER, Dissertation. Utrecht 1 Maart 1921.

The heart has served us as a type of the two automaticities; the natural one depending on the radio-activity of potassium, or rubidium, and the artificial one, that can be evoked by the radio-activity of uranium, thorium, ionium, radium, emanation. In quite the same way there is a mutual resemblance between the alpha- and beta-automaticities of the gut and the uterus.

Botany. — *"Further researches on the antiphototropic curvatures occurring in the coleoptiles of Avena."* By Dr. C. E. B. BREMEKAMP. (Communicated by Prof. F. A. F. C. WENT.)

(Communicated at the meeting of May 27, 1922).

As I have shown in my former communication [1]), the conditions under which the coleoptiles of Avena produce an antiphototropic curvature, may be summed up in this way:

1st. At the end of the one-sided illumination the rate of growth should have about the same value at both sides of the coleoptile [2]). This result is only to be obtained with light of rather strong intensity. If this is provided for, the product of the intensity and the exposition-time should exceed a certain value.

2nd. After the close of the illumination, there should be a more rapid increase of the growth-rate, in the side that has received the greatest quantity of light. In this way it should reach here a higher value.

An explanation of the way wherein a difference in the rate of increase may come about, was given in my paper entitled: "Theorie des Phototropismus" [3]). After a previous diminution in consequence of the illumination, the rate of growth after some time increases again. This process commences probably the sooner, according as the diminution has been the greater. In this way the increase of the growth-rate in the side which has received the greatest quantity

---

[1]) C. E. B. BREMEKAMP. On antiphototropic curvatures occurring in the coleoptiles of Avena. Proceedings Kon. Akad. v. Wetensch. te Amsterdam. Vol. XXIV, p. 177. 1921.

[2]) In my previous work in stead of the expression "the rate of growth at the end of the illumination" I used the ampler expression "the rate of growth belonging to the grade of sensibility existing at the end of the illumination". In this way I reckoned with the possibility that it would give a latent period between the phototropical reaction i.e. the change of the rate of growth. and the absorption of the light with its influence on the sensibility. However, a critical examination of the literature on this subject, has convinced me that the evidence in favour of the existence of this latent period, is not conclusive. The investigations of BOSE and others have made it very probable that the reaction follows the illumination almost immediately.

[3]) C. E. B. BREMEKAMP. Theorie des Phototropismus. Rec. d. trav. bot. Néerlandais Vol. XV. p. 123. 1918.

of light, may gain an advantage of that in the other side. This advantage will be the greater, according as there lies more time between the moment whereon the growth-rate in the anterior side has reached its lowest value, and the moment whereon this is the case in the posterior side. If it is sufficiently great, the rate of growth in the first-named side with the aid of it will reach at the end of the illumination or shortly afterwards a higher value. In any case the exposition-time should be long enough that an advantage of sufficient extent may be gained.

However, in my previous communication I showed that an antiphototropic curvature may come about also, if the expositiontime is very short. As in this case the explanation given above naturally fails, I suggested that the theory of Bose [1]) might give us here the clue to get out of the difficulty.

According to this theory, a disturbance of equilibrium in the organism generally manifests itself in a local contraction (the direct effect) which is accompanied by an expansion in the adjoining tissue (the indirect effect). In the latter, the turgescence would be heightened by the water expelled from the contracted portion, and accordingly a temporary enhancement of the growth-rate would be the result. In this way a normal curvature in one part of an organ would always go together with an antitropic one in the adjoining region. Only if an increase of the rate of growth in that part, should be impossible, the antitropic curvature would remain out. In our case then, the origin of the antiphototropic curvature in the tip of the coleoptile might be connected with the origin of a normal phototropic curvature in the basal part.

To test the correctness of this supposition, I made a number of experiments wherein the phototropic reaction of coleoptiles exposed in the whole of their length, was compared with the reaction of coleptiles illuminated at the tip only, or illuminated also in the whole of their length, but after an exposition of the basal part to a two-sided illumination of rather great strength.

Before I enter into the details of these experiments, I will give a survey of the results which previous investigators have obtained in their researches on the influence of an illumination of one part, on the phototropic reaction of another.

First of all then, we have to consider the experiments on phototropism made by Bose [2]) himself. They are rather few in number,

[1]) J. C. Bose. Plant Respose. London 1906.

[2]) J. C. Bose assisted by Jyotiprakash Sircar. The transmitted effect of photic stimulation. Life Movements in Plants. Calcutta 1918/19. p. 362—377.

and form only a subordinate part in the general frame of his work. His experimental objects were seedlings of Setaria and roots of Sinapis.

The choice of the first-named object is not very happy, as the direct effect of the exposition of the coleoptile is not outwardly visible, and its existence therefore, as yet purely hypothetical. The indirect effect consists in an antitropic curvature of the axis. This curvature which appears almost immediately, is followed in about 25 minutes by a normal one. The latter should be the result of the propagation of the invisible direct effect. An illumination of the growing region gives a normal curvature.

That the antitropic curvature of the axis occurring with an exposition of the coleoptile should be the indirect effect of this illumination in the sense of Bose, is possible. It should be remarked however, that it is not proved. As yet, we don't know with certainty, if in this case the direct effect consists really in a contraction, as no sign thereof becomes outwardly visible.

The roots of Sinapis show a negative phototropism. At least this is the case, when both the tip and the growing region are exposed to the light. The curvature appears in the growing region, the tip always remaining straight. An exposition of the tip also gives a negative curvature of the growing region, but if this part itself is exposed to the light, there appears at first a positive curvature which only after some time is followed by a weak negative one.

Bose considers the negative curvature in the growing region produced by an exposition of the tip, as the indirect effect, the direct effect as in Setaria remaining concealed. That this curvature is not, as in Setaria, followed by a positive one, he explains by assuming that the intervening tissue would be practically unable to conduct the direct effect. In the case of an exposition of the growing region, the positive curvature is considered as the direct effect, whereas the negative curvature appearing a little later, is said to arise on account of transverse conduction of the direct effect under continued illumination.

However, this explanation is not very convincing. That a neutralisation of the curvature might come about by transverse conduction, is quite conceivable, but how a reversion of the curvature might be explained in this way, I fail to understand. Moreover, as a conductivity for the direct effect in the longitudinal direction is supposed to be absent, it is not readily admissible that is should be very efficacious in the transverse direction. Therefore, in this case the interpretation of Bose cannot be considered as sufficiently founded.

The explanations of these negative curvatures given by other investigators are, however, hardly more convincing.

Information about the influence of an illumination of the basal part on the reaction of the tip, is to be found in papers by VAN DER WOLK [1]), GUTTENBERG [2]) and ARISZ [3]), all dealing with the phototropism of Avena.

According to VAN DER WOLK the results of an illumination of the basal half of the coleoptile on the upper half, is to be seen in the fact, that an illumination of 12 MCS gives in these seedlings a curvature of the same strength as an illumination of 85 MCS in a wholly etiolated coleoptile. This greater curvability of the upper half might, perhaps, be explained by assuming that the contraction of the tissue in this part was facilitated by the decrease of turgescence in the basal half: the expulsion of the water would find here less resistance.

GUTTENBERG on the contrary, tried to show, that the curvability of the tip of the coleoptile is not altered by an illumination of the basal part. In his experiments three sets of seedlings were compared. They were all illuminated unilaterally with 22,2 or 33,3 MCS; but in the second and third set the basal part was exposed previously during one hour to an illumination with 11,1 MC; in the second set the seedlings rotated during this time round a vertical axis, whereas in the third set they stood still. In this case the after-illumination took place from the opposite side. GUTTENBERG found that the phototropic curvature in the third set was a little weaker than in the other two.

This result seems at first in flagrant contradiction to the statement of VAN DER WOLK cited above, but it should be remembered that in the experiments of VAN DER WOLK, the light was very strong, and the exposition only short, whereas GUTTENBERG used light of rather feeble intensity and a very long exposition. Therefore, in the seedlings of VAN DER WOLK the decrease of turgescence in the basal part, might have been greater, and consequently the effect on the curvability of the tip more important than in the seedlings of GUTTENBERG. This explanation would probably suffice, if there was no difference at all between the curvatures in the three sets. GUT-

---

[1]) P. G. VAN DER WOLK. Investigations of the transmission of light stimuli in the seedlings of Avena. These Proceedings, Vol. XIV, p. 327.

[2]) H RITTER VON GUTTENBERG. Ueber akropetale Reizleiting. Jhrb. f. wis. Bot. Bd. 52 p. 333. 1913.

[3]) W. H. ARISZ. Untersuchungen über den Phototropismus. Rec. d trav. bot. Néerlandais. Vol. XII p. 44. 1915.

TENBERG stated however, that the curvature in the third set was smaller than in the other two, and explained this discrepancy by assuming a propagation of the basal curvature to the tip. In my opinion it might have its cause in the circumstance, that these seedlings were already slightly curved at the moment of the after-illumination. If this had been the case, the tip would have received here a smaller quantity of light and this moreover partly under a less favourable angle than in the other sets, and consequently, the phototropic curvature would not have attained the same value. It should also be mentioned, that a repetition of these experiments by ARISZ (l.c. p. 105) gave only doubtful results, the sources of error being very great. In any case, we dare not say, that the acropetal propagation of the basal curvature has been demonstrated, and for the solution of the question, whether an exposition of the basal part exercises any influence on the curvability of the tip, the experiments are not suitable, the intensity of the light being too weak.

Nevertheless, there are in the paper of GUTTENBERG a few indications, which seem to show that the illumination of the basal part influences the curvability of the tip, in the way described by VAN DER WOLK. On p. 341 one may read: "Ein deutlicher Unterschied zwischen den Krümmungswinkeln der beiden Serien war dabei nicht zu konstatieren; doch verhielten sich die allseits vorbeleuchteten Pflanzen zunächst etwas anders als die verdunkelten. Bei ersteren erfährt nämlich das oberste Drittel der Koleoptile eine etwas stärkere Krümmung als bei letzteren, dafür ist aber bei diesen die Krümmung bereits weiter nach unten fortgeschritten". That these differences were only very small (further data l.c. p. 437) and quantitatively very different from those observed by VAN DER WOLK, may find its explanation, as I have pointed out already, in the feeble intensity of GUTTENBERG's illumination.

ARISZ mentions (l. c. p. 103), that he has repeated the experiments of VAN DER WOLK, and describes his results in this way: „Wohl ist in vielen Fällen eine kleine Vergrösserung der Spitzenkrümmung beobachtet worden, welche auch etwas früher sichtbar wurde, aber so eklatant, wie VAN DER WOLK seine Resultate beschreibt, war es nicht". ARISZ therefore does not deny, that the illumination of the basal part enhances the curvability of the tip; only he awards this influence less importance than VAN DER WOLK does.

Summing up, we may state that our knowledge of the influence which an exposition of the basal part exercises on the tip, is far from complete. Moreover, it cannot be said that the available data

are very valuable for our supposition, that the antiphototropic curvatures of Avena might find their explanation in this way.

In the experiments of ARISZ (l.c. p. 97), the exposition of the basal part gave a normal curvature [1]), which did not extend itself beyond the limits of the part exposed. As the occurrence of an antiphototropic curvature in the tip is never mentioned, we must assume that under the circumstances of these experiments, the tip remained perfectly straight. At first sight, this seems to clash with our supposition, but we should remember, that in these experiments the tip remained continually in the dark, so that its turgescence underwent no decrease. Now in consequence of this circumstance, an increase of the rate of growth might be difficult or even impossible.

In my own experiments I compared in the first place the reaction of coleoptiles exposed at the tip only, with the reaction of coleoptiles exposed in the whole of their length. The result was very clear. Whereas in the first case antitropic curvatures were never found, in the second case they could be obtained without difficulty.

The etiolated seedlings used for these experiments, were planted in a single row in oblong zinc boxes. Each box got about 15 seedlings, so orientated, that their plane of symmetry was parallel to the small side of the box. During the exposition, the boxes were placed perpendicular to the rays of light. The seedlings that should be exposed at the tip only, stood with their basal part behind a screen, so that only $2\frac{1}{2}$—3 mm. of the tip protruded. This screen was prepared in the following way. A feeble red light was placed just in front of the experimental lamp, and the silhouette of the coleoptiles caught on a piece of black paste-board standing just behind them. The place of the tip was marked thereon with the aid of a pencil. Above these marks the paste-board was cut away and then the screen pushed $2^1/_2$—3 mm. deeper in the earth. After that the box was turned round and the red light removed. During the illumination with the experimental lamp, in this way just $2^1/_2$—3 mm. of the tip was exposed.

The intensity of the illumination was in all experiments 750 MC;

---

[1]) In two experiments out of a very great number, ARISZ mentions to have obtained antitropic curvatures in the part exposed. In one case (illumination during 1 minute with 330 MC), the curvatures are stated to have been feebly normal or antitropic, in the other case (illumination during 1 minute with 200 MC), they were antitropic or absent. As these cases, however, stand wholly isolated among the rest of his results, it seems probable that these antitropic curvatures are due to some experimental error.

the exposition-time 12, 15, 18 and 21 seconds. The temperature varied between 15° and 20° C., but in each series of experiments it remained nearly constant. After the illumination the boxes came on the clinostat.

With an exposition of 12 seconds (light-quantity 9000 MCS), after $3^1/_2$ hours the coleoptiles exposed in the whole of their length, were feebly antitropic (S-shaped), the coleoptiles exposed at the tip only, feebly curved in the normal way.

With an exposition of 15 seconds (light-quantity 11250 MCS), the results were nearly the same.

With an exposition of 18 seconds (light-quantity 13500 MCS), after $3^1/_2$ hours the coleoptiles exposed totally, were clearly antitropic (feebly S-shaped), the coleoptiles exposed at the tip only, nearly straight.

With an exposition of 21 seconds (light-quantity 15750 MCS), after $3^1/_2$ hours the coleoptiles were all nearly straight.

The experiment with the exposition-time of 15 seconds, was repeated 5 times, always with the same result. That in this case, the occurrence of an antitropic curvature at the tip of the totally exposed coleoptiles, is dependant upon the exposition of the basal part, cannot be doubted.

The results of the experiments wherein the basal part of the coleoptiles was previously exposed to a very strong illumination, and where, therefore, the unilateral after-illumination of the whole coleoptile did not give a normal curvature in the basal part, demonstrate the significance of this influence also clearly.

In these experiments, I compared the result of an unilateral illumination of the whole coleoptile after a twosided exposition of the basal part, with that of an unilateral illumination of seedlings previously kept in the dark. During the fore-illumination two screens of the same shape were used, one in front of the coleoptiles, and one behind them. They were prepared in the same way as those used in the previous experiments, the only difference being that in this case, the basal part of the paste-board was for the greater part cut away. In this way during the illumination, at the tip of the coleoptiles a piece of $2^1/_2$—3 mm. remained in the dark. The fore-illumination lasted 60 seconds, and during this time, every 10 seconds the box was turned round. At the end of the fore-illumination the box was turned round for the last time, then the screens ware taken away, and the seedlings once more exposed to the same light. This time the illumination lasted 12 or 15 seconds. The intensity of the illumination was always 750 MC. The result of these experiments

was, that the coleoptiles, whereof the basal part was previously exposed, remained straight, whereas the others showed the usual antiphototropic curvature.

In my former communication I admitted that in coleoptiles previously submitted to an omnilateral illumination of a definite value, an antiphototropic curvature might, perhaps, be obtained with the aid of a rather weak after-illumination. This seems now not very probable, as under these circumstances, the occurrence of a normal curvature in the basal part, may hardly be expected. Therefore, in this case neither of the causes hitherto discovered, by which an antiphototropic curvature may be produced, is present.

The relative importance of the two causes is as yet wholly unknown, but that the cause discussed in this paper, must be very efficient, follows from the experiments described in my former communication (l.c. p. 182). The antiphototropic curvatures produced by an illumination with a given quantity of light, showed but little difference if the exposition-time varied between 1 and 256 or between $^1/_4$ and 192 seconds. Now, as we have seen that with a very short exposition-time, the presence of the cause discussed in my earlier work, is wholly excluded, we must conclude that its influence in the experiments with a longer exposition, was here also rather weak.

SUMMARY.

The antiphototropic curvature which appears at the tip of the coleoptile of Avena with a very short exposition, does not show itself, if the illumination is limited to the tip, or if the basal part has previously been exposed to a rather strong illumination.

Therefore we should assume, that with an unilateral illumination of the whole coleoptile, the rate of growth of the tip, is enhanced by an influence proceeding from the basal part. This influence must be greatest in the side, which underwent the greatest contraction, that is to say in the side, which during the exposition faced the lamp. The origin of an antiphototropic curvature of this kind is, therefore, always connected with the origin of a normal curvature in the basal part.

Mathematics. — *„Ueber Wirkungsfunktionen"*. By Prof. R. WEITZENBÖCK. (Communicated by Prof. L. E. J. BROUWER).

(Communicated at the meeting of May 27, 1922.)

§ 1. *Einleitung.*

Bei der Ableitung der Feldgesetze und der Erhaltungssätze in der allgemeinen Relativitätstheorie und deren Erweiterungen steht man vor folgender Aufgabe: wenn g_{ik} und φ_i die Komponenten eines Kovarianten Tensors. 2. resp. 1. Stufe sind und

$$g_{ik,\alpha} = \frac{\partial g_{ik}}{\partial x_\alpha}, \quad g_{ik,\alpha\beta} = \frac{\partial^2 g_{ik}}{\partial x_\alpha \, \partial x_\beta}, \quad \varphi_{i,\alpha} = \frac{\partial \varphi_i}{\partial x_\alpha}, \quad \varphi_{i,\alpha\beta} = \frac{\partial^2 \varphi_i}{\partial x_\alpha \, \partial x_\beta} \quad . \quad (1)$$

gesetzt wird, so ist aus diesen Funktionen eine absolute Invariante W zu bilden. $W\sqrt{g}$ wird dann eine relative Differentialinvariante vom Gewichte eins (= eine scalare Dichte) und

$$\int \mathfrak{W} \, dx = \int W \sqrt{g} \, dx = \iiiint W \sqrt{g} \, dx_1 \, dx_2 \, dx_3 \, dx_4 \quad . \quad . \quad (2)$$

wird eine absolute Integralinvariante.

Man nennt $\mathfrak{W}$ die Wirkungsfunktion. Bedeutet δ eine Variation der g_{ik} und φ_i, so gibt die Gleichung

$$\delta \int \mathfrak{W} \, dx \equiv 0 \quad \text{(für alle } \delta g_{ik} \text{ und } \delta \varphi_i) \quad . \quad . \quad . \quad . \quad (3)$$

die Feldgesetze.

Die Frage nach allen Differentialinvarianten zweiter Ordnung der beiden Tensonen g_{ik} und φ_i wird zurückgeführt auf die einfachere Frage nach allen ganzen, rationalen Differentialinvarianten dieser Tensoren. Hierauf gibt ein Reduktionssatz von RICCI und LEVI-CIVITÀ [1]) die Antwort: man hat alle projektiven Invarianten der folgenden 5 Tensonen zu suchen:

$$\left.\begin{array}{ll} g_{ik} & = \text{metrischer Fundamentaltensor} \\ \varphi_i & = \text{electromagnetisches Potential} \\ R_{ik,\alpha\beta} & = \text{(Riemann-Christoffel'scher) Krümmungstensor} \\ \varphi_{i(\alpha)} & = \text{erste kovariante Ableitung der } \varphi_i \\ \varphi_{i(\alpha)(\beta)} & = \text{zweite kovariante Ableitung der } \varphi_i. \end{array}\right\} \quad . \quad (4)$$

[1]) Mathem. Ann. 54, (1901), p. 138.

Die Frage nach *allen* projektiven Invarianten dieser Tensoren bildet ein sehr kompliziertes algebraïsches Problem. (Nach dem allgemeinen Endlichkeitssatz von HILBERT gibt es *endlich-viele* ganze rationale Invarianten, durch die sich alle übrigen ganz und rational ausdrücken lassen.)

Glücklicherweise ist hier die Sache nicht so trostlos verwickelt, indem zwei sehr einschränkende Forderungen gestellt werden: in der EINSTEIN'schen Theorie wird verlangt, dass die Feldgesetze Differentialgleichungen höchstens zweiter Ordnung werden; in der Theorie von WEYL müssen die aus den Tensoren (4) gebildeten Wirkungsfunktionen auch masstabsinvariant sein.

Wir behandeln zuerst den zweiten Fall.

§ 2. *Die Theorie von* WEYL.

In der durch WEYL gegebenen Erweiterung der allgemeiner Relativitätstheorie muss die aus den Tensoren (4) gebildete Wirkungsfunktion absolut-invariant gegenüber Masstabstransformationen sein. Diese Transformationen sind gegeben durch

$$g'_{ik} = \lambda g_{ik} \qquad \varphi'_i = \varphi_i - \frac{\partial \log \lambda}{\partial x_i}, \quad . \quad . \quad . \quad . \quad (5)$$

woraus noch entsprechende Gleichungen für $R'_{ik,\alpha\beta}$, $\varphi'_{i(\alpha)}$ und $\varphi'_{i(\alpha)(\beta)}$ folgen.

Die Forderung $\mathfrak{W}' = \mathfrak{W}$ (für alle λ) erniedrigt dann die Anzahl 5 der Tensoren (4) auf 4 masstabinvariante Tensoren:

$$\left.\begin{array}{ll} g_{ik} & = \text{metrischer Fundamentaltensor} \quad (g'_{ik} = \lambda\, g_{ik}) \\ f_{ik} & = \text{electromagnetisches Feld} \quad (f'_{ik} = f_{ik}) \\ {}^*F_{ik,\alpha\beta} & = \text{Richtungskrümmung} \quad ({}^*F'_{ik,\alpha\beta} = \lambda\, {}^*F_{ik,\alpha\beta}) \\ E_{ik,\alpha} & = f_{ik(\alpha)} - \tfrac{1}{2}\,(f_{\alpha k}\varphi_i + f_{i\alpha}\varphi_k + 2 f_{ik}\varphi_\alpha - g_{\alpha i} f_{\beta k}\varphi^\beta - g_{\alpha k} f_{i\beta}\varphi^\beta) \\ & \qquad (E'_{ik,\alpha} = E_{ik,\alpha}). \end{array}\right\} \quad . \quad (6)$$

Der gegenüber $\mathfrak{W} = W\sqrt{g}$ etwas allgemeinere Ansatz $\mathfrak{W} = W g^h$, wobei W keinen Faktor g mehr enthält, führt weiters auf die Gleichung

$$2n_2 + 2n_3 + 3n_4 = 4,$$

wobei W ganz und rational vom Grade n_2, n_3, n_4 in den f_{ik}, ${}^*F_{ik,\alpha\beta}$ und $E_{ik,m}$ ist. Daher ist $n_4 = 0$ und für n_2 und n_3 bleiben nur die drei Möglichkeiten (2,0), (1,1) und (0,2) übrig. Man kann dann beweisen[1]), dass sich unter diesen Annahmen nur die folgenden sechs Wirkungsfunktionen ergeben:

[1]) R. WEITZENBÖCK, Wiener Ber. 129, (1920), p. 683 und p. 697; dito, 130, (1921), p. 15.

$$\left.\begin{aligned}
\mathfrak{W}_1 &= \sum_{ik} f_{ik} f_{i'k'} = 2(f_{12}f_{34} + f_{13}f_{42} + f_{14}f_{23}) \\
\mathfrak{W}_2 &= f_{ik} f^{ik} \sqrt{g} \\
\mathfrak{W}_3 &= \frac{1}{\sqrt{g}} \sum_{ik} \sum_{lm} {}^*F_{ik,lm} {}^*F_{i'k',l'm'} \\
\mathfrak{W}_4 &= \sum_{ik} {}^*F^{oo,\lambda\mu}_{ik,oo} {}^*F_{i'k',\lambda\mu} \\
\mathfrak{W}_5 &= \left({}^*F^{oo,ik}_{ik,oo}\right)^2 \sqrt{g} \\
\mathfrak{W}_6 &= {}^*F^{ik,lm} {}^*F_{ik,lm} \sqrt{g}
\end{aligned}\right\} \quad . \quad . \quad . \quad (7)$$

Hiezu machen wir die folgenden Bemerkungen[1]). $\mathfrak{W}_1$ und $\mathfrak{W}_4$ kommen als Wirkingsfunktionen nicht in Betracht, da ihre Variationen identisch Null geben, wie R. Bach bewiesen hat[2]). $\mathfrak{W}_2$ ist die Maxwell'sche Wirkingsfunktion, bei Weyl mit $\mathfrak{l}$ bezeichnet[3]). Auch $\mathfrak{W}_5 = F^2 \sqrt{g}$ wird von Weyl verwendet.

An Stelle von $\mathfrak{W}_3$ kann man auch die Invariante

$$W_3' = {}^*F_{ik} {}^*F^{ik} \quad , \quad \mathfrak{W}_3' = W_3' \sqrt{g} \quad . \quad . \quad . \quad . \quad . \quad (8)$$

verwenden; es ist nämlich:

$$\mathfrak{W}_3 = -\mathfrak{W}_3' + \tfrac{1}{4}\mathfrak{W}_2 + \tfrac{1}{4}\mathfrak{W}_5 + \mathfrak{W}_6 \quad . \quad . \quad . \quad . \quad . \quad (9)$$

Die Variationen von $\mathfrak{W}_3'$ und $\mathfrak{W}_6$ wurden von W. Pauli[4]) und R. Bach[5]) berechnet.

§ 3. *Die Theorie von* Einstein.

In der Einstein'schen Theorie ist $\mathfrak{W} = W\sqrt{g}$ und W ist aus den Tensoren (4) zusammen gesetzt: rational in den g_{ik}, ganz und rational in den übrigen vier Tensoren.

Variieren wir die g_{ik} allein, so bekommen wir die Gravitationsgleichungen $\mathfrak{W}^{ik} = 0$; die Variation von φ_i ergibt die verallgemeinerten Maxwell'schen Gleichungen $\mathfrak{w}^i = 0$. Dabei sind diese „Tensordichten"[5]) gegeben durch:

$$\mathfrak{W}^{ik} = \frac{\partial \mathfrak{W}}{\partial g_{ik}} - \frac{\partial}{\partial x_\alpha}\left(\frac{\partial \mathfrak{W}}{\partial g_{ik,\alpha}}\right) + \frac{\partial^2}{\partial x_\alpha \partial x_\beta}\left(\frac{\partial \mathfrak{W}}{\partial g_{ik,\alpha\beta}}\right) . \quad . \quad . \quad (10)$$

$$\mathfrak{w}^i = \frac{\partial \mathfrak{W}}{\partial \varphi_i} - \frac{\partial}{\partial x_\alpha}\left(\frac{\partial \mathfrak{W}}{\partial \varphi_{i,\alpha}}\right) + \frac{\partial^2}{\partial x_\alpha \partial x_\beta}\left(\frac{\partial \mathfrak{W}}{\partial \varphi_{i,\alpha\beta}}\right) . \quad . \quad . \quad (11)$$

---

[1]) H. Weyl, Phys. Zeitschr., 22, (1921), p. 473.

[2]) R. Bach, Mathem. Zeitschr. 9, (1921), p. 124.

[3]) H Weyl, Raum, Zeit, Materie, 4. Aufl., (1921), p. 268.

[4]) W. Pauli, Phys. Zeitschr., 20, (1919), p. 457; Verhdl. d. Deutsch. Phys. Ges. 21, (1919), p. 742.

[5]) D. Hilbert, Göttinger Nachr. 20. 11. 1915.
R. Weitzenböck, Wiener Ber. 130, (1921), p. 15.

Berechnet man diese „Variations-Ableitungen" und verlangt man, dass sie Differentialquotienten von höchstens zweiter Ordnung enthalten, so ergeben sich die drei folgenden Möglichkeiten:

A. W enthält die $R_{ik,\alpha\beta}$ linear, keine $\varphi_{i(\alpha)}$ und keine $\varphi_{i(\alpha)(\beta)}$:

$$W = A\,(g_{ik}\ ,\ \varphi_i\ ,\ R_{ik,\alpha\beta})\,; \quad . \quad . \quad . \quad . \quad . \quad . \quad . \quad (12)$$

B. W enthält die $\varphi_{i(\alpha)(\beta)}$ linear, keine $R_{ik,\alpha\beta}$ und keine $\varphi_{i(\alpha)}$:

$$W = B\,(g_{ik}\ ,\ \varphi_i\ ,\ \varphi_{i\,(\alpha)\,(\beta)})\,; \quad . \quad . \quad . \quad . \quad . \quad . \quad (13)$$

C. W enthält keine $R_{ik,\alpha\beta}$ und keine $\varphi_{i(\alpha)(\beta)}$:

$$W = C\,(g_{ik}\ ,\ \varphi_i\ ,\ \varphi_{i(\alpha)})\,. \quad . \quad . \quad . \quad . \quad . \quad . \quad . \quad (14)$$

Wir behandeln diese drei Fälle der Reihe nach. Bei A kann man zeigen, dass man nur die zwei Invarianten erhält:

$$A_1 = R = g^{ik} R_{ik} \quad , \quad A_2 = R_{ik}\,\varphi^i\,\varphi^k \quad . \quad . \quad . \quad . \quad (15)$$

A_1 ist das von Einstein verwendete R.

Im Falle B haben wir drei Invarianten:

$$B_1 = \varphi^{\ i}_{o\,(\alpha)\,(i)}\,\varphi^\alpha \quad , \quad B_2 = \varphi^{o\,\alpha\,o}_{i\,o\,(\alpha)}\,\varphi^i \quad , \quad B_3 = \varphi_{i\,(\alpha)\,(\beta)}\,\varphi^i\,\varphi^\alpha\,\varphi^\beta. \quad (16)$$

Die neben B_1 noch mögliche Invariante

$$B'_1 = \varphi^{\ i}_{o(i)(\alpha)}\,\varphi^\alpha$$

ist mit Hilfe von B_1 und A_2 ausdrückbar:

$$B_1 - B'_1 = \varphi^{\ i}_{o\,(\alpha)\,(i)}\,\varphi^\alpha - \varphi^{\ i}_{o\,(i)\,(\alpha)}\,\varphi^\alpha = -R_{\alpha\beta}\,\varphi^\alpha\,\varphi^\beta = -A_2 \quad (17)$$

Komplizierter ist der dritte Fall C. Hier ist die Anzahl der Invarianten sehr gross: das Aufsuchen aller Invarianten kommt hinaus auf das Berechnen eines vollen Systems von orthogonalen Invarianten einer quaternären Linearform φ_i und einer ebensolchen (unsymmetrischen) Bilinearform $\varphi_{i\,(\alpha)}$. Dies ist eine bisher noch ungelöste Aufgabe.

Wir führen einige der einfachsten Invarianten vom Typus C an. Enthält C erstens keine $\varphi_{i\,(\alpha)}$, so haben wir die einzige Invariante

$$C_1 = \varphi = \varphi_i\,\varphi^i = g^{ik}\,\varphi_i\,\varphi_k \quad . \quad . \quad . \quad . \quad . \quad . \quad (18)$$

Wenn C die $\varphi_{i(\alpha)}$ linear enthält, haben wir zwei Invarianten

$$C_2 = \varphi_{i(\alpha)}\,\varphi^i\,\varphi^\alpha \quad , \quad C_3 = \varphi^{\ i}_{o\,(i)} = \frac{1}{\sqrt{g}}\,\frac{\partial\,(\varphi^i\sqrt{g})}{\partial x_i} \quad . \quad . \quad (19)$$

Die Wirkungsfunktion $C_3\sqrt{g}$ gibt zu den Feldgesetzen keinen Beitrag, da $C_3\sqrt{g}$ eine Divergenz ist.

Von den in den $\varphi_{i\,\alpha)}$ quadratischen Invarianten C nennen wir nur noch

$$C_4 = 2\,(\varphi_{i(\alpha)}\,\varphi^{i(\alpha)} - \varphi_{i(\alpha)}\,\varphi^{\alpha(i)}) = f_{ik}\,f^{ik}; \quad . \quad . \quad . \quad . \quad (20)$$

Hier ist f_{ik} das elektromagnetische Feld und $C_4\sqrt{g}$ ist die MAXWELL'sche Wirkungsfunktion.

Sind $f_\sigma(\varphi)$ Polynome von φ (Vgl. (18)) mit constanten Koeffizienten, so hat die allgemeinste Wirkungsfunktion die Gestalt

$$\mathfrak{W} = [f_0(\varphi)+f_1(\varphi)A_1+f_2(\varphi)A_2+f_3(\varphi)B_1+f_4(\varphi)B_2+f_5(\varphi)B_3+C]\sqrt{g} \quad (21)$$

C bedeutet hier eine ganze rationale Funktion von Invarianten (14).

Von dieser Wirkungsfunktion ausgehend, wären nun die Feldgesetze aufzustellen. Dies ist bisher nur für die einfachsten Invarianten durchgeführt worden.

Bacteriology. — "*Studies on the bacteriophagus of* d'Herelle". By J. W. Janzen and L. K. Wolff. (Communicated by Prof. C. Eykman).

(Communicated at the meeting of May 27, 1922).

IV. *About the relation between bacteriophagus and resistant bacteria.*

d'Herelle tells us in his book that, when a weak bacteriophagus is added to a thick emulsion of bacteria, the former will have disappeared from the suspension after some time.

Then he says that the bacteriophagus also seems to penetrate into the bacteria, but that, now that the bacilli could not increase, the bacterium resists the bacteriophagus, which is destroyed in vivo. We have considered it important to study this phenomon carefully once more; for this we have used some typhoidbacteriophagi, one resistant and one not resistant typhoidstrain out of our collection.

We have found that the disappearance of the bacteriophagus as described by d'Herelle for thick emulsions also takes place in the ordinary thin emulsions, this time not of normal but of resistant bacilli.

We have also found that old non resistant bacilli, which are not being dissolved by the bacteriophagus in consequence of their age, do absorb the latter; in this case however, the bacteriophagus only increases when the bacteria multiplicate and so get young again.

Some of the series of experiments about this subject are as follows:

Series of experiments I.

Resistant strain = T 20. Non resistant strain = T Wi.

Determination of the number of bacteriophagus germs by counting the number of islands (on agarplate).

Bacteriophagus Wi,

Adding equal portions of bacteriophagus Wi to equally turbid suspension in broth of T 20 and T Wi.

Number of bacteriophagus germs per cM^3.

	T 20	T Wi.
Directly	(18 milliard)	18 milliard
1/4 hour	0.6 „	0.28 „
3/4 hour	2.4 „	innumerable
1 1/2 hours	0 8 „	„
24 hours	0.02 „	

A second experiment with T (Sm) instead of T (Wi) offered an analogous result.

	T 20	T Sm.
Directly	(30 milliard)	30 milliard
after 1/4 hour	1.7 „	4 „
„ 1 hour	4 „	100 „
„ 24 hours	1.8 „	innumerable

After a week the number of bacteriophagus germs with the resistant strain was about the same as the number found after 24 hours; with the non resistant strain it had greatly increased.

We have regularly found the slight increase (in comparison with the number after 1/4 hour and after 3/4—1 hour) with the resistant strain; the explanation seems to us as follows: the resistant strain also has some weaker descendants which can be dissolved by the bacteriophagus; hence an increase of the bacteriophagus, which is now being caught by the stronger brothers.

We can easily succeed in destroying the bacteriophagus by culturing three of four times on broth with new bacilli the mixture of bacteriophagus-resistant strains in fresh broth.

I. 1st culture of bacteriophagus germs	60 milliard per cM^3
2nd „ „ „ „	24 „ „ „
3rd „ „ „ „	1/4 „ „ „
4th „ „ „ „	disappeared?
5th „ „ „ „	disappeared

II.	1st culture of bacteriophagus germs	18 milliard per cM³
	2nd „ „ „	0 02 „ „ „
	3rd „ „ „	disappeared

Finally an experiment with old non resistant bacilli.
A. 14 days old bacilli *Sm* in broth.
B. 6 hours old bacilli *Sm* in broth.

A.	B.
Directly (30 milliard)	30 milliard
after 1/4 hour 4 „	6 „
after 3/4 „ 2 „	800 „
after 2 „ 32000 „	Innumerable.

Our typhoid bacilli that are resistant by nature to the bacteriophagus do not even lose their resistance after being subcultured repeatedly in contradistinction to what D'Herelle tells us in his book (pag. 67) about the bacilli, who are been made resistant by influence of the bacteriophagus.

V. About big and small islands.

O. Bail and T. Watanabe have said that, in plating a mixture of bacteriophagus and bacteriacultures on agarplates, the islands are not always equally big, but that sometimes big ones, medium ones and small ones are to be found.

They have tried to cultivate the bacteriophagus of these islands purely; they say that they have succeeded in doing this with the small islands, not with the big ones however.

We too had already been struck by this before Bail's communication reached us, and we have tried to isolate these bacteriophagi, forming big and small islands, from each other, but we did not succeed. We have stated though, that it could not be possible in our cases, as a bacteriophagus which exclusively formed big islands with regard to one typhoidstrain, made nothing but small islands with regard to another typhoidstrain, and as to a third, both big and small ones. So we do not believe that Bail's and Watanabe's explanation is right, but we think the difference in size of the

islands must be attributed to a difference of virulence as to the various strains. Big islands point to a strong effect with regard to the typhoidstrain; small ones to a weaker effect. This has also been proved by a still to be published investigation of Dr. KROPVELD in our laboratory, about staphylococci-bacteriophagi.

Bacteriophagus W i always gives both big and small islands with regard to T W i.

Small and big islands are cultured over separatly 9 times, small islands always being used for the series of small ones, big islands for the series of big ones in this process.

The last culture of both always gave a mixture of big and small islands again.

Finally we have tried both bacteriophagi W i big [10] and W i small [10] as to 4 typhoidstrains.

With both bacteriophagusstrains we got exactly the same result which is only following once.

1. Clearing.	2. Checking.	3. Islandformation.
Typhoid 9 ++++	++++	++++ very big islands.
„ 17 +++	++++	++++ big islands.
„ 27 —	±	++++ very small islands.
„ Wi ++++	++++	++++ big and small islands.

Laboratorium of Hygiene.

Amsterdam, May 1922.

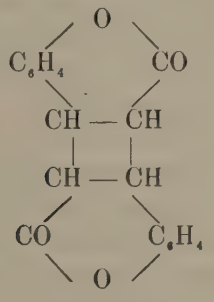
Chemistry. — *"The Biscoumaric Acids"*. By A. W. K. DE JONG.

(Communicated at the meeting of May 27, 1922).

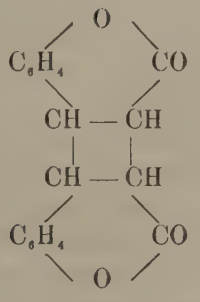
Some time ago [1]) I communicated that the product of illumination of coumarin is not identical with hydrodicoumarin of FITTIG and DYSON, as CLAMICIAN and SILBER had thought, but that it must have another structure, because when treated with alkalis it does not give a mono-basic, but a di-basic acid.

It is natural to suppose that the product of illumination of coumarin is formed from coumarin in the same way as α- and β-truxillic acid are formed from the forms of normal cinnamic acid by the combination with formation of a tetramethylene ring between the doubly bound C-atoms of the two molecules.

As two molecules of normal cinnamic acid can combine in four different ways to a truxillic acid [2]) also the combination of two molecules of coumarin will give four different biscoumarins, which will, as the truxillic acids, belong to two series according to the arrangement of the C-atoms with unequal (I) or equal (II) atom-groups next to each other in the tetramethylene ring.



I.



II.

Of both structural formulae two different biscoumarins can exist according to the situation of the coumarinrings on different sides or on the same side of the tetramethylene ring.

---

[1]) These proceedings Vol. XX, 875.

[2]) These proceedings Vol. XX, 590.

To the product of illumination of coumarin one of these four structural formulae must be assigned.

Also another biscoumarin is known, obtained by KNUT T. STRÖM [1]) by boiling biscoumaric acid, formed by illumination of coumaric acid, with anhydrous acetic acid. This biscoumarin is, as STRÖM already communicated, different from the biscoumarin obtained by illumination of coumarin, nor is it identical with the hydrodicoumarin of FITTIG and DYSON.

The biscoumaric acid of STRÖM is formed from coumaric acid, of which no metastable forms are known till now, in a conformable way as α-truxillic acid of α-normal cinnamic acid, and therefore it is very likely that this biscoumaric acid will have a conformable structure to α-truxillic acid. The properties of this biscoumaric acid known at present are in agreement with this, as will be shown.

The biscoumarin of STRÖM would then possess the structual formula I, the coumarin-rings being situated on different sides of the ring.

To distinguish the different biscoumaric acids I propose to give to these acids similar names as to the truxillic acids, and then the biscoumaric acid of STRÖM must be called α-biscoumaric acid, and its biscoumarin α-biscoumarin. The melting- at the same time decomposition-points of the two substances are the same, viz. 318° (STRÖM stated them to be above 275°); α-biscoumaric acid also changes into its biscoumarin when heated to 250°. The biscoumarin obtained by illumination of coumarin might be different from α-biscoumarin by the position of its coumarin-rings situated, on the same side of the tetramethylene-ring or it might be one of the two other possible biscoumarins indicated by figure II. The first supposition was not very likely, the two biscoumarins showing no change when heated at 210° with the acetic acid anhydride, whilst, when they had only a difference in the situation of the coumarin-rings with respect to the tetramethylene-ring, a change of one into the other was probable. This experiment is, however, not a conclusive proof of a different binding of the coumarin-molecules in the biscoumarins. The best way to decide this is to prepare the acid of the biscoumarin, converting it to the dimethylether, and to try if through heating with the acetic acid anhydride at 210° an anhydride is formed which gives a dimethylether of another biscoumaric acid. If the two coumarin-rings are situated on the same side of the tetramethylene ring, no other biscoumaric acid is formed, whilst

---

[1]) Ber. **37**, 1383.

when they are on different sides, a new biscoumaric acid will be obtained.

The methylation of α-biscoumaric acid by dimethylsulfate gives the dimethylester of the dimethylether crystallized into needles, melting at 133° and sparingly soluble in ether. On boiling with alkalis the dimethylether was obtained melting at 261°—262°. BERTRAM and KÜRSTEN [1]) found the melting point of this substance, obtained by illumination of the methylether of coumaric acid, to be 260—262°.

When the dimethylether is heated with the acetic acid anhydride at 210°, the anhydride of the dimethylether of γ-biscoumaric acid was formed, which crystallized in pretty large bright yellow crystals out of the anhydride of acetic acid, melting at 186°—187°. The dimethylether itself was obtained in fine needles melting at 234°.

When the α-biscoumaric acid is heated with KOH the acid corresponding to β cocaic acid was obtained, which separated in an ether solution by addition of petrolether in rhomb-shaped crystals melting 212°. As it whould be strange to give this acid a name connected with coca, I propose to call it ζ-biscoumaric acid. With a similar treatment also the dimethylether of α-biscumaric acid gave the same acid, which shows that the methylgroups are split off through melting with KOH

These transformations of the α-biscoumaric acid, respectively the dimethylether, are wholly analogous to these of α-truxillic acid.

The dimethylester of the dimethylether of the biscoumaric acid of the product of illumination of coumarin, for which I propose the name of λ-biscoumaric acid, melts at 112°—113°; the dimethylether itself at 134°.

By heating the dimethylether with acetic acid anhydride at 210° and after evaporating the solvent in a glycerine bath at about 130° a brown sirup was obtained, which did not crystallize. The acid obtained by boiling the sirup with alkali crystallizes out of an etherpetrolether solution in fine needles melting at 203°. On account of its resemblance in structure with ε truxillic acid I propose to call this substance the dimethylether of ε biscoumaric acid. This transformation proves that the coumarin-rings of the illumination product are situated on different sides of the tetramethylene ring and as also α-biscoumarin possesses the same situation of the coumarin-rings and the two substances are different, λ-biscoumarin must have the structure of fig. II and by the removing of a carboxylgroup

[1]) Journ. f. pr. Ch. (2) **51**, 323.

from one side of the tetramethylene ring to another an o-dioxy-ε-truxillic acid is formed.

By melting with KOH λ-biscoumaric acid is converted into δ-biscoumaric acid, crystallizing in needles melting at 157°.

I hope to make further communications on other possible transformations of the biscoumaric acids, while it will also be tried to obtain them from the truxillic acids, by which the proposed names and the structural formulae will obtain more security.

Laboratory of the Colonial Museum, Haarlem.

Physics. — "*On the Excitation and Ionization Potentials of Neon and Argon*". By G. HERTZ. (Communicated by Prof. P. EHRENFEST).

(Communicated at the meeting of May 27, 1922).

It is known that rare gases and metallic vapours behave in a very simple way on collision with slow electrons. Then there can be exchange of energy between electrons and atoms only in one way, viz. that in which the transferred energy is used to bring the colliding atom into a higher quantum condition. Hence on collision with the atoms the electrons can transfer only very definite energy quanta to them, which according to BOHR's theory, are in direct connection with the series-spectrum of the atom. For a great many metallic vapours this transition of energy in quanta has already been investigated and the relation to the optic spectra has been shown. Of the rare gases accurate measurements have only been carried out for helium [1]), on the ground of which FRANCK succeeded in making the system of the series-spectra of helium complete, and in showing the connection between the ortho-helium and the par-helium spectrum. Several observations have, indeed, been made for neon and argon [2]), but the results are inaccurate for the greater part, and partly in conflict with each other. Besides in the great sensitiveness of noble gases to traces of impurities, the excitation and ionization potentials of which lie nearly always below that of the rare gas, the cause of these conflicting results seems to lie chiefly in this that the efficiency of the unelastic collisions in the rare gases is much smaller than in the metallic vapours, so that the methods which lead to good results for the latter, cannot be applied here. In order to attain reliable results, it seemed, therefore, necessary to me, to refine the methods for the investigation of the quantum

[1]) F. HORTON and A. C. DAVIES, Proc. Roy. Soc. London (A) **95**, 408, 1919.
J. FRANCK and P. KNIPPING, Zeitschr. f. Physik, **1**, 320, 1920.
K. T. COMPTON, Phil. Mag. **40**, 553, 1920.

[2]) F. HORTON and A. C. DAVIES, Proc. Roy. Soc. London, (A), **97**, 1, 1920 and **98**, 124, 1920.
G. STEAD and P. S. GOSLING, Phil. Mag. **40**, 413, 1920.
H. C. RENTSCHLER, Phys. Rev. **14**, 503, 1913.
G. DÉJARDIN, C. R. **172**, 1347, 1921.

transition of energy between electrons and atoms, and supplement it by a method which admits of a clear distinction between light emission and ionization also in the case of unelastic collisions of small efficiency.

The methods applied up to now for the study of the quantum emission of energy consist in this that either the radiation or ionization that take place starting from a definite potential, or the phenomenon that the impinging electrons lose energy, is used as a proof of the occurrence of unelastic collisions. In this way a curve is obtained in which the different steps of energy appear as breaks; an accurate measurement of them is often difficult, especially for the higher steps of energy. It seemed, therefore, desirable to me to use as criterion for the quantum transition of energy a characteristic that immediately disappears again when the critical potential is exceeded, and consequently causes the separate steps of energy to stand forth as sharp maxima. Such a characteristic is the occurrence of electrons with the velocity zero. For as soon as an electron possesses exactly the energy required for the excitation of a definite quantum transition, it may lose all its energy at the collision, and be left behind as an electron with the velocity zero. If, however, it possesses a greater energy, it retains the rest after the collision, and remains behind as an electron with a velocity which, though smaller, is yet different from zero. If, therefore, electrons of a definite velocity, are admitted into a space in which they collide with atoms of a noble gas, electrons of the velocity zero will only occur when the energy of the electrons is precisely equal to the work required for the excitation of a quantum transition. When, therefore, the number of electrons which are left behind with the velocity zero, is plotted as function of the accelerating potential, a sharp maximum must be obtained for every potential corresponding with an energy-quantum that can be transferred at a collision of electrons. In consequence of the inevitable distribution of velocity of the electrons it is not possible to determine the number of electrons which have rigorously a velocity zero. Therefore the number of those electrons the velocity of which lies below a definite small value (in our measurements mostly 0.2 Volt) will be plotted as function of the tension.

The measurements according to this principle are carried out in the following way:

The electrons emitted by a short incandescent wire D of tungsten (Fig. 1) enter the field-free space R through the gauze N_1 after acceleration through an electric field, in which space they collide with the atoms of a rare gas. Part of the electrons passes through

the cylindrical gauze N_2 after numerous collisions; opposite this gauze a receiving plate has been adjusted, also cylindrical. (The cylindrical arrangement of N_2 and P appeared to be preferable, though good results were also obtained with an apparatus with two parallel pieces of gauze and a plane receiving plate). When between N_2 and P a small retarding potential is applied, all the electrons, the velocity of which corresponds to smaller potentials, are held back. A certain part of the faster electrons will likewise be retained by the weak counter-field, but as appears on closer consideration, this part greatly decreases with increasing velocity. The difference between the stream of electrons received on the plate with and without the small counterfield, therefore, gives a measure for the number of electrons having about the velocity zero. In order to be able to measure this difference with great accuracy, an arrangement was chosen which rendered it possible to insert and cut out the field alternately; the part of the potentiometer from which the small counter-potential had been branched off, could be short-circuited by a mercury contact in vacuum. By alternate reading of the deviation with and without counterfield the difference could be accurately read, and an error in consequence of a possible change of the zero-point was out of the question. It is further possible to render oneself independent of a slow change in the emission of electrons of the incandescent wire, by dividing the difference measured by the total deviation. For the efficiency of the method it is of importance that the metal surfaces should have the greatest purity possible, as small impurities can already cause Volta-potential differences of the order of magnitude of the small counter potential. The copper used had been cauterized with nitric acid immediately before the construction and the sealing in of the apparatus. The whole apparatus was mounted on a pretty large glass foot, as is used for incandescent lamps, so that it could be fused into a glass globe without the metal parts being heated too much. It was heated at 400° in high vacuum for six hours; after this the copper, even though it was a little tarnished before in a few places, presented a perfectly pure metallic surface.

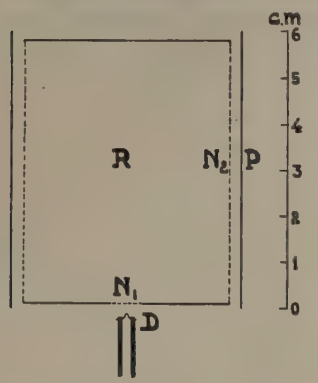


Fig. 1.

In figures 2 to 4 curves are represented as instances of the results of such measurements, which refer to neon of a pressure of 0,51 m.m., to a neon-helium mixture of 30% helium and a pressure

of 0,56 m.m. and argon of 0,36 m.m. pressure. Especially fig. 3 shows the efficiency of the method. In spite of the comparatively small percentage of helium, the two first excitation tensions of helium, though they lie above the strong excitation tensions of neon,

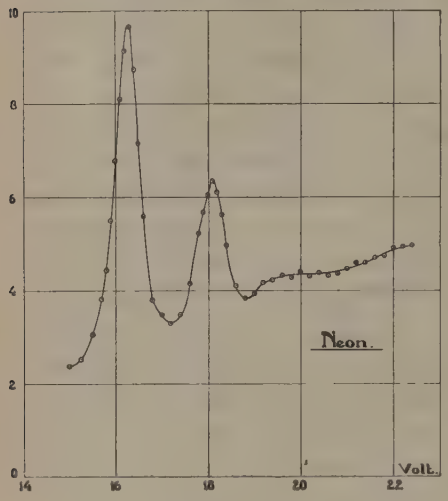


Fig. 2.

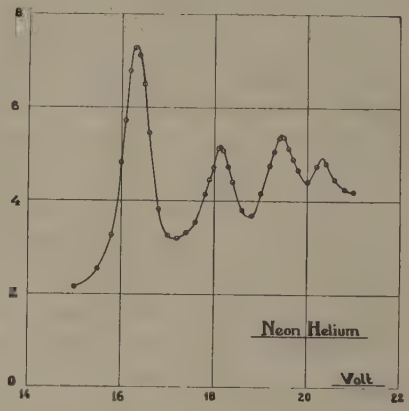


Fig. 3.

stand out as two sharp maxima at a distance of 0,8 Volt. These maxima were used to obtain the absolute value of the excitation tensions of neon, in which the value of 20,45 Volts measured by FRANCK and KNIPPING[1]) for the lowest excitation tension of helium, was used as basis. The values thus obtained appeared to be independent to a high degree of the circumstances of the experiment.

This method is entirely unsuitable for the measurement of the ionization-potential. For then the impinging electron or the electron that has been liberated from the atom by the collision can have the velocity zero after an ionizing collision, also when the energy of the colliding electron was greater than the work of ionization. As moreover at first the effect brought about by ionizing collision evidently rapidly increases with increasing tension, the curve shows no maximum here, but only a rise, which is besides influenced by the positive ions, and does not admit an accurate determination of the ionization-potential. For this reason the arrangement usual with a strong counter-field between N_2 and P was used for the measurement of the tension of ionisation for some measurements; most observations were, however, made according to a new method,

[1]) J. FRANCK and P. KNIPPING, l.c.

in which a very reliable criterion is applied for the first appearance of the ionization. For this purpose a second, very thin incandescent wire G of the shape represented in Fig. 5 was placed in the field-

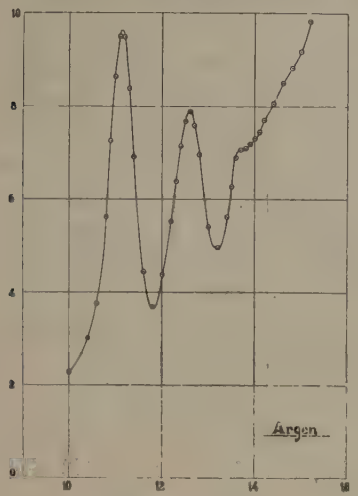


Fig. 4.

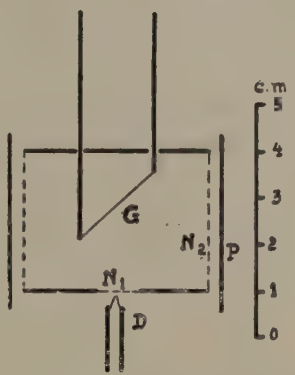


Fig. 5.

free space R; the positive end of this wire (on the left side of the figure) was connected with the walls of R, so that the field round the wire, which moreover remains restricted to its immediate neighbourhood on account of the slight thickness of the wire, can by no means accelerate the electrons coming from D. This incandescent wire was heated to such a temperature that the stream of electrons flowing from the wire to the metal wall, is limited by the space charge. So long as the energy of the electrons coming from D is not sufficient for the formation of positive ions, they have no influence at all on the amount of the stream of electrons issuing from G. Nor could a photo-electric effect, if it took place, even apart from the fact that it is so small, have any influence on the amount of this stream of electrons, limited by the space charge. As soon, however, as positive ions are formed, and some of them get into the neighbourhood of G, the space charge is partly annihilated, and the stream starting from G suddenly rises. Figures 6 and 7 show the results of these measurements in neon and argon. It is seen that not even the slightest discontinuity can be perceived in the curve when the lower excitation tensions are passed, while at the tension of ionisation the stream begins to rise rapidly. To obtain the absolute value of the ionization potential, the maximum corresponding to the first excitation potential was determined at the same time by the aid

of N_2 and P according to the method discussed above, which is likewise expressed in the figures. In this it should be taken into

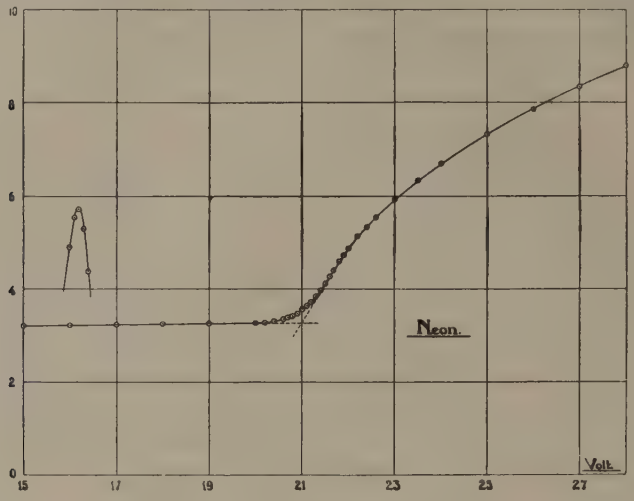


Fig. 6.

account that this maximum slightly differs with respect to the tension at which unelastic collisions take place, viz. the amount equal to the small counter-potential (here 0,2 Volt). It may also be mentioned that the measured stream in these experiments was about

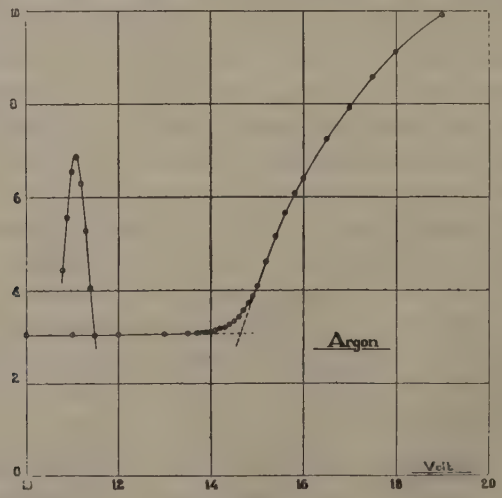


Fig. 7.

thirty times the flow of electrons issuing from D, so that in this way an accurate measurement of the ionization potential is possible, even with an ordinary millivoltmeter.

Results. Starting from the value of 20,45 Volts for the lowest excitation potential of helium, two rather pronounced excitation potentials were found for neon at 17,35 and 19,15 Volts, 22,2 Volts was found for the ionization-potential of neon; for argon two excitation potentials were found at 12,25 and 13,7 Volts, a less distinct one at 14,7 Volts; the ionization-potential at 16,0 Volts.

With the very complicated structure of the optical spectrum of neon the occurrence of discrete fairly pronounced excitation potentials seems surprising at first sight. If, however, the serial scheme of neon drawn up by PASCHEN [1]) (fig. 8) is plotted in a scheme in the way given by BOHR, it is directly seen that the values found are in very good harmony with the optical measurement. The term

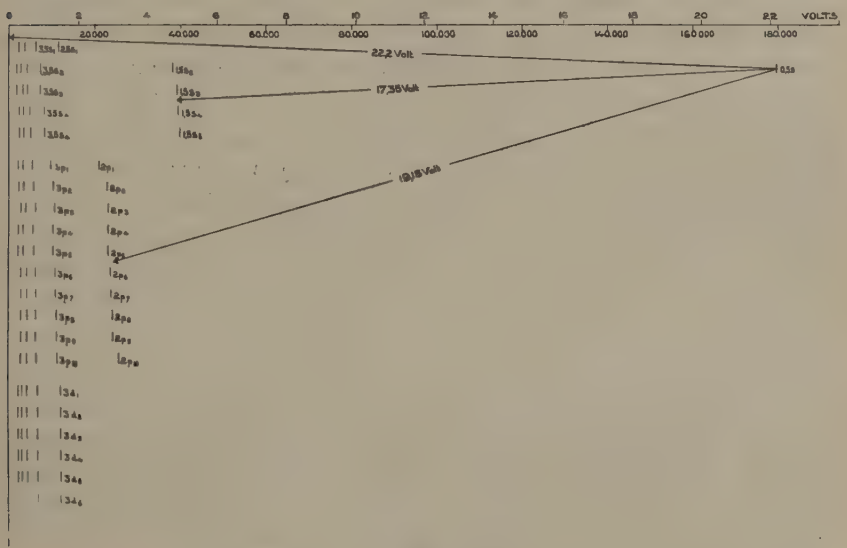


Fig. 8.

corresponding with the normal state and denoted by me as $0{,}5s$ has been added in the scheme, the value of this term has been calculated from the ionization-potential. This term $0.5\,s$ is first of all followed by a group of four terms of the type of $1.5\,s$; these lie close together within a region which, expressed in Volts, is smaller than 0,2 Volt., and can, therefore, not be separated in measurements with colliding electrons. Then follows a group of $2\,p$ terms, the greater part of which lies again within 0.1 Volt. After this come, about 1 Volt higher, the $3d$ terms. The other terms succeed each other at distances of at most some hundredths of Volts,

[1]) F. PASCHEN, Ann. d. Phys. **60**, 405, 1919.

so that the methods of the collisions of electrons is not sufficient to separate them, only a "continuous spectrum" can be observed. When the curve given in fig. 3 is compared with this, the serial system is clearly found back in it. The first maximum corresponds to the group of the quantum transitions $0.5\,s$—$1{,}5s$, the second to the group $0.5\,s$—$2\,p$, and then at a distance of 1 Volt follows the spectrum of the transitions to higher quantum conditions, which seems continuous on account of the small dissolving power. Also quantitative the agreement is good, as is seen in fig. 8, where the quantum transitions observed with collisions of electrons have been indicated by arrows, of which the projection on the axis of abscissae is equal to the observed value of the potentials of excitation resp. ionization. It is, therefore, seen that the serial scheme of neon has become complete by the addition of the term $0.5\,s = 179800 \pm 1000$. There is no room in this serial system for a resonance-potential of 11.8 Volts and ionization-potentials at 16.7 and 20 Volts, which values were derived by HORTON and DAVIES [1]) from their experiments, nor was there any indication at all in my measurements of the occurrence of resonance or ionisation at these potentials. On the other hand the experiments of the same investigators on excitation of light in neon through collision of electrons [2]) are in good agreement with the conclusions which may be derived from the completed scheme. As can at once be read from the figure, the lines of the principal series must first appear alone starting from 19.2 Volts, then from about 20.2 Volts the lines of the secondary series must gradually begin to make their appearance, while the whole spectrum only can be emitted above the ionization-potential. HORTON and DAVIES actually found that at 20 Volts only the lines of the principal series were emitted, the whole spectrum not appearing before 22.8 Volts.

It would be of importance to ascertain whether there may perhaps be terms for neon that correspond to metastable states, as FRANCK has found them for helium and mercury [3]). To find this out it would, however, be necessary really to separate the different terms, and for this the dissolving power of the method of the collisions of the electrons is not yet sufficient. Measurements with the usual arrangement for showing photo-electric radiation proved, as was to be expected, the occurrence of photo-electrically active radiation at both the excitation potentials observed.

As was already stated above, in *argon* there likewise appear two

[1]) F. HORTON and A. C. DAVIES, l.c.

[2]) F. HORTON and A. C. DAVIES, Phil. Mag. **41**, 921, 1921.

[3]) J. FRANCK, Phys. Zeitschr. **22**, 388, 1921.

excitation potentials (at 12.25 and 13.7 Volts), and a less distinct one at 14.7 Volts, which is followed by a series of energy steps which has not yet been dissolved. Here too the apparently sharp excitation potentials will no doubt correspond with undissolved groups of terms lying close together, on account of the complication of the argon spectrum. The argon spectrum not yet having been split up into series, a comparison is not yet possible. When a similar structure is assumed for the spectrum of argon as for neon, then starting from the measured values for excitation and ionization potentials, the following mean values for the first groups of terms are to be expected:

$$0.5\ s = 130000 \pm 1000$$
$$1.5\ s = 30400$$
$$2\ p = 18600$$
$$\text{higher terms} < 10500$$

The serial terms calculated by NISSEN [1]) do not fit in with this scheme. Also the fact that according to NISSEN lines of the red and the blue argon spectra are considered as members of the same series, though the condition for the excitation of the two spectra are different, pleads in my opinion against the validity of the terms calculated by him.

For the rest more complications may possibly be expected for argon than for neon. The fact found by PASCHEN [2]) that for part of the neon series the limits are shifted by a constant amount in comparison with the other series, was explained by GROTRIAN [3]) by the aid of the L-doublet of neon. He has also already pointed out that it must be expected for argon thad the multiple M-limits will manifest themselves in an analogous way.

Physical Laboratory of the "N. V. Philips Gloeilampenfabrieken".
(Philips Incandescent Lamp Works).
Eindhoven.

---

[1]) K. A. NISSEN, Phys. Zeitschr. **21**, 25, 1920.
[2]) F. PASCHEN, Ann. d. Phys. **63**, 201, 1920.
[3]) W. GROTRIAN, Zeitschr. f. Phys. **8**, 116, 1921.

Microbiology. *"On the Occurrence of Sulphate-reduction in the deeper layers of the Earth".* By C. A. H. VON WOLZOGEN KÜHR. (Communicated by Prof. G. VAN ITERSON JR).

(Communicated at the meeting of April 29, 1922).

§ 1. *Introduction.*

The disappearance of organic matter at greater depths in the soil has since long occupied the minds of investigators. The difficulties associated with their inquiries regard especially that of obtaining sterile samples from such depths, which is essential to microbiological inquiry.

The process of oxidation, which causes organic matter to disappear, can be effected by free as well as by combined oxygen. When the air is shut off, as is the case in the lower strata, oxidation is of course brought about by combined oxygen.

Now the question is how this process can take place microbiologically.

The term sulphate-reduction designates the process by which, with the exclusion of air, organic matter in the soil is oxidized under the influence of combined sulphate-oxygen. This anaerobic process is effected by Microspira desulfuricans, discovered in 1895 by BEIJERINCK [1]). It being an exothermic process energy is set free through this oxidation, which is utilized physiologically by the sulphate reducing spirilla. The rough equation for sulphate-reduction gives the following formula:

$$2C\ldots + CaSO_4 + H_2O \rightarrow CaCO_3 + CO_2 + H_2S,$$

in which C... is the symbol for the source of carbon.

Microspira desulfuricans occurs in the mud of ditches and the ooze of the Dutch "Wadden". The grey, bluish-black to black colour of the soils in which sulphate-reduction takes place, must be ascribed to ferric sulphid, in which form the liberated hydrogen sulphid is combined by iron-compounds present in the soil.

The occurrence of the sulphate-reducing microbe at the greater depths in the terrestrial soil has been less frequently observed and,

---

[1]) Ueber Spirillum desulfuricans als Ursache von Sulfatreduktion. Verzamelde Geschriften. 3de deel, pg. 102.

to my knowledge, statements about it are few and far between. JENTZSCH [1]) e.g. records that in the deeper ooze-layers of the ocean of about 40 m. and more, reduction-processes occur, in which hydrogen sulphid and ferric sulphid are formed, which are ascribed by him to decomposition of proteins. It is more likely, however, that here also we have to do with sulphate-reduction, since it has been proved that this is of frequent occurrence under the circumstances alluded to.

Another statement is given by EUG. DUBOIS [2]) who observed the transformation of sulphate into ferric sulphid in the lower alluvial clay-layers underneath the Dutch Dunes.

An opportunity to ascertain the occurrence of sulphate reduction in deeper layers was offered, when in the autumn of 1921 a number of new wells were dug along the Sprenkelkanaal on the source of supply of the Amsterdam Dune Waterworks.

§ 2. *How the samples of sand, clay and peat were obtained from the well-shafts.*

In connection with the bacteriological sampling it will be well to set forth, in principle, the way in which the new wells were sunk.

A wide iron tube is driven vertically into a dug, shallow cavity. The sand is excavated from a greater depth than is at first reached by the tube, which can consequently sink gradually deeper. By means of a screw-thread one length of tube is screwed on to the other, so that a system of tubes is procured of the length necessary to reach a certain depth.

The masses of sand and the occasional lumps of clay and peat are removed from the tubes with a so-called "puls", consisting of a hollow iron cylinder of smaller diameter than the tube's. At the lower end it is sharp-edged to facilitate the sinking, while the bottom is provided with a valve, opening to the inside. By means of two iron bars that are fastened to the edge of the open top-part of the cylinder and are suspended on the same point of support, it is possible to connect the apparatus to a pulley-block. When moving the "puls" forcibly up and down in the wet mass of sand present in the shaft, it is ultimately filled with a pap of sand. The filled "puls" is then hoisted up and emptied by overturning it. This process of removing the sand from the well-shaft is briefly called "pulsen".

---

[1]) Zeitschrift d. Geol. Ges. 1902. 54, p. 144. Cf. RAMANN, Bodenkunde, p. 180.

[2]) Het Leidsche Duinwater. Eene hydrologische studie. 1912, p. 19 en 20.

§ 3. *The examination of the sand- and clay-samples for sulphate-reduction.*

From a chemico-biological point of view it is interesting to ascertain the origin of the ferric sulphid, which gives a dull-grey, greyish blue to bluish-black colour to the soil-samples. The obvious hypothesis that the ferric sulphid was formed by sulphate-reduction, was in every respect substantiated by the examination of the many sand-, and clay-samples procured by means of the "puls". Thus the sulphate reduction in the deeper layers of the earth underneath the dunes appeared to be a bacteriological process of common occurrence.

The demonstration of sulphate reducing spirilla was performed after the accumulation-method of BEIJERINCK, the culture-medium [1]) used being:

Tapwater	100	
Na-lactate	0.5	
Asparagin	0.1	
$MgSO_4 . 7$ aq.	0.05	(or gypsum)
$FeSO_4 . 7$ aq.	0.001	

with which sterile stoppered bottles of ± 150 cc. capacity were filled after infection with a quantity of the sand-, or clay-samples under examination. They were filled up to the neck, then cautiously stoppered and placed under 25° C.

BEIJERINCK [2]) showed that in this anaerobic procedure Microspira desulfuricans is exclusively the causative agent of the sulphate-reduction manifesting itself, as appears from the formation of hydrogen sulphid and the black ferric sulphid.

My culture bottles showed in every respect the same progress of the reduction process, so that hereby the examined sand-, and clay-samples gave evidence of the presence of Microspira desulfuricans.

The material used for infection of the medium was drawn from the inner portion of the sand-mass in the "puls" by means of a sterile spatula, and deposited in sterile wide-mouthed stoppered bottles. Directly when the samples were received at the laboratory they were subjected to investigation.

Sterile sampling could be effected to perfection only in clay-, and peat-samples. This was performed after BEIJERINCK's [3]) method. The sample was split in two. From the fracture laid bare, the required

---

[1]) A. VAN DELDEN. Beitrage z. Kenntn. d. Sulfaatreduktion durch Bakt. Centralbl. f. Bakt. 2e Abt. 1903. Bd. XI, p. 83.

[2]) Verzamelde Geschriften. (Collected Papers) Vol. 4, p. 53.

[3]) Verzamelde Geschriften. (Collected Papers) Vol. 2, p 354. Note 2.

inoculation material was taken by means of a sterile spatula. The lumps of clay and peat suited our purpose well, since in the splitting the fracture was not contaminated by crumbling particles of the edges, which was owing to the solid structure of the samples resulting from their humidity.

The time in which the formation of hydrogen sulphid in the culture-bottles commenced was very different for the same inoculation-substance and especially for the sulphate reduction it largely depends on the number of viable germs present at the outset of the experiment.

The clay-, and the peat-samples dredged up with the "puls", were derived from the clay-, and the peat-banks underlying the dunes. They were all compact masses, in which the original stratified structure, arising from sedimentation, had been preserved. These clay-, and peat-layers being all but impermeable to water, their inside represents the original bacteriological condition of the stratum, from which the sample has been taken.

The clay- and peat-lumps were on the outside wet and on the inside, judging superficially at least, moderately humid. The water-content of the clay amounted to about 26%; in the clay-samples which contained peat in the stratified structure, the content of moisture was considerably higher, viz. about 50%. The peat-samples exhibited the largest amount of water, viz. rather more than 77%.

The clay- and the peat-lumps varied from very large ones to those of the size of a fist and appeared to meet the bacteriological requirements in every respect.

§ 4. *Summary of results and observations on the inquiry about sulphate-reduction.*

The number of soil-samples of the 9 wells which were examined for sulphate-reduction, have been summarized in the subjoined table.

The quantum of infection-material used for every sulphatereduction-test amounted to from 5 to 10 grs. of the soil-sample. After an interval of from 3 to 20 days sulphate-reduction revealed itself at 25° C., which period rose to 5 weeks in the case of the peat-sample B 31.

In every well, even the deepest of 34.50 m. below A.P., we chiefly found sand over the whole depth, in which irregularly spread lens-shaped clay-, and peat-layers occurred alternately.

With a few exceptions all the sand- and clay-samples indicated in the subjoined table, yielded on examination for sulphate-reduction a conclusive positive result. Consequently the dull-grey or grey colour

of the sand-samples and the mostly blue to bluish-black colour of the clay-samples points to prevailing sulphate-reduction. This com-

B 22	B 24	B 25	B 26	B 27	B 28	B 29	B 30	B 31
21.6 M.	35 34 M.	35.30 M.	6.50 M.	*8.00* M.		6.50 to 10.50 M.	*9.50* M. 12.50 »	*8.00* M. *14.00* »
			15.10 » 17.50 »	*13.25*°) »	*6.00* M. 6.50 »	14.00 to 16.25 »	18.50 » 32.50 »	25.30 » (peat)
			28.50 »		16.50 »	*20.50*°) »	34.50 »	
			32.50 »		*29.00* »	*23.10* »		
					34.50 »			

B 22, B 24, etc. = wells.

The values express in metres the depths below A.P. (= Amsterdam level) from which the soil samples have been drawn.

The figures in italics refer to clay-samples which enclose organic particles or peat-layers.

The figures in ordinary type are sand-samples.

(°) = no sulphate reduction in culture bottle.

mences at about 10 m. below the surface (7.5 m. —A. P.)[1] to ± 37 m. (34.50 m.—A.P.) the largest depth examined here.

The conditions under which sulphate-reduction appears are:

1°. Absence of oxygen [2].

2°. The occurrence of organic compounds.

3°. The presence of sulphate and the required mineral compounds.

The first condition, the absence of oxygen, is satisfied in consequence of the considerable depth below the level of the ground.

The second condition: the occurrence of organic compounds, is fulfilled already to the eye by the peat-sample and also by the clay-sample with enclosed peat-layers. That the sand-, and clay-samples, which do not enclose immediately distinguishable organic particles, also contain organic matter, can be demonstrated chemically, by the potassiumpermanganate method. This is conducted as follows: The soil-sample is boiled with diluted sulphuric acid and filtered. The filtrate is cooled down under the tap; now potassiumpermanganate (0.01 norm.) is instilled. The first drops are directly decolorized. which is owing to the oxidation of ferro- and mangano-compounds,

[1]) The grounds of the wells at the Sprenkelkanaal is lying at 2.5 M. above A. P.

[2]) Traces of oxygen are left out of consideration here.

Then a moment follows in which the colour of the added potassium-permanganate disappears only slowly: this is the oxidation of the organic matter, extracted by the diluted sulphuric acid, for in a drop of this extract, placed on a piece of filterpaper soaked with potassium ferrocyanid no ferro can be demonstrated any more.

The sand-samples are most often not so rich in organic compounds as the clay-samples, which often contain peat. Presumably this generates a stronger sulphate-reduction than is possible in the sand-samples, and this is probably the reason why clay can be darker in colour than sand.

VAN DELDEN [1]) has shown that for sulphate-reduction organic bodies are required which are easily oxidizable. This justifies the assumption that in the organic substances, demonstrated by us, there are some bodies difficult of oxidation and others again which are easily oxidizable, which is proved indirectly by the sulphate-reduction that manifests itself in the sand-, clay-, and peat-samples.

Also the 3rd condition, the presence of the required mineral compounds, was satisfied. In our examination for sulphate only small amounts could be demonstrated, which is explained by the disappearance of sulphate through sulphate-reduction.

One of the mineral combinations is that of the insoluble, black-coloured ferric sulphid, formed by the iron and the liberated hydrogen sulphid, as pointed out already in § 3.

From the foregoing we may deduce that the conditions of anaerobic life which we found in the deeper layers of the soil, fairly agree with the prevailing sulphate-reduction.

§ 5. *The content of „aerobic" and „anaerobic" germs of the deeper layers of the soil.*

Besides the demonstration of sulphate-reducing spirilla in the soil-samples, another question arises, viz. whether they contain other germs and whether these belong to the aerobes or the anaerobes. We examined the samples:

B 28 29.00 M — A.P. (clay with peat)
B 29 6.50 — 10.50 M — A.P. (clay).
B 31 25.30 M — A.P. (peat).

The number of germs was ascertained in the way described in § 3. With a sterilized spatula inoculation-material was taken from the soil-samples, it was then shaken up in sterile tapwater and

[1]) Centralbl. f. Bakt. Bd. XI, 2te Abt. 1903, p. 83.

subsequently weighed. This material was used for making counting-tests by sowing the micro-organisms on nutrient gelatin. The counting of the microbe-colonies for the aerobic plate-cultures took place after 48 and 72 hours, after which there was hardly any increase of the colonies worth mentioning.

The anaerobic culture plates for the counting-tests were made after WRIGHT and BURRI's [1]) culture method, modified by me. As this strictly anaerobic method of cultivation yields very good results, it will not be amiss to state our procedure.

In a glass box closed tightly by a glass stopper with a ground rim a smaller petri-dish is placed containing a solidified culture-medium on which the anaerobes are sown in streaks. The circular open space left round the dish is first stopped up with non-absorbent cotton-wool on which a layer of absorbent cotton-wool is laid. The latter is soaked with 20 % potassium hydrate and finally with an equal volume of 20 % pyrogallic acid.

Throughout this procedure the petri-dish remains covered. After the cotton-wool has been soaked with pyrogallic acid the dishcover is removed, while the glassbox is closed by its cover-glass of which the glass-rim is smeared with vaselin. The rim of the glass-box may also be shut off with paraffin after the lid has been adjusted. In order to facilitate the opening of the glass-boxes, the wall is provided with a little hole which is shut off with paraffin and is opened again before taking off the lid of the box, in

Soil-sample.	Aerobes.		Anaerobes.	
	number of germs per c.c. of soil.		number of germs per c.c. of soil.	
B 28 29.00 M. — A.P. clay + peat.	After 48 hrs.	After 72 hrs.	After 4 days.	After 12 days.
	15400	20000	818	409000
B 29 6.50—10.50 M. — A.P. clay.	20	130	—	400
B 31 25.30 M. — A.P. peat.	—	7000	103600	160000

[1]) J. H. WRIGHT. A method for cultivation of anaerobic bacteria. Centralbl. f. Bakt. 1te Abt. 29, 1901, pg. 61. R. BURRI. 2te Abt. 1902. 8, pg. 533.

order to admit the air. Now the cover of the petri dish is easily removed.

The number of anaerobes was counted in the same way as that of the aerobes in the same sample.

Because we had determined the specific weight of the soil-samples, we could establish the number of germs per cc.

Our results we have tabulated on page 7.

The time in which the anaerobes yielded a constant number of colonies was considerably longer than that of the aerobes.

It strikes us that the anaerobic test yields a total of germs which is much greater than that of the aerobic one, while the amount of germs in B 28 and B 31 is much higher than that of B 29. The last-named fact is perhaps due to the higher content of organic matter in the first two soil-samples.

For the sake of comparison we may add that in raw water from the dunes the number of bacteria per c.c. varies in round numbers from 400 to 1800.

§ 6. *It appears that microbes derived from aerobic and anaerobic cultivation belong for the greater part to the facultative anaerobes.*

The number of species of bacteria obtained in the preceding paragraph by the method described, appeared to be only small when we examined their qualities. Generally the anaerobes and the aerobes [1]) were not identical. The following table shows the number of species of microbes we found:

Soil-sample.	Aerobes.	Anaerobes.
B 28 29.00 M. — A.P.	2 species	4 species
B 29 6.50 — 10.50 M. — A P.	2 »	1 »
B 31 25.30 M. — A.P.	1 »	4 »

As to their properties aerobes revealed some resemblance in acidformation from glucose, Berlin-blue formation from ferri-ferricyanid, the formation of hydrogen-sulphid from broth (lead-carbonate test), the splitting of aesculin, the formation of katalase, and most often in the inability to ferment glucose, to form lipase and diastase. Spores were not formed.

[1]) Probably B 29 anaerobe and one of the species B 29 aerobe were identic.

A difference in the properties of the two microbe-groups appeared from the following reactions: Anaerobes form nitrite from nitrate in a marked degree, indican is split extensively in most cases (oxidation of indoxyl to indigo-blue), a moderate amount of invertase is formed, a large amount of slime (wall-matter) is formed from saccharose. Aerobes lack these qualities. They liquefy gelatin, whereas the anaerobes do not.

My investigation into the properties of the microbes did not put me in a position to classify them.

When examining microbes derived from aerobic cultivation for their anaerobic behaviour, it appeared that only B 31 grew very well without air, those of B 28 and B 29, however, very badly. The occurrence of these aerobes seems to show that presumably very small quantities of air are to be found at larger depths in the soil, and that they are carried along with the rainwater that penetrates at a very slow rate into the deeper layers of the earth. If the layer, as is the case here with clay, is only sparingly permeable to water, the dissolved oxygen is allowed to diffuse to the places where it is to be consumed.

The microbes obtained from anaerobic cultivation developed enormously when living in air. This appeared conclusively when the anaerobic culture-boxes after being opened had been standing for some time exposed to the air. Then the microbe colonies grew larger and larger in a very short time. These bacteria grew very well as aerobes, also on nutrient agar-slants. Tested in this way the majority of the isolated bacteria appeared to belong to the *facultative anaerobes*, which is consistent with the occurrence of these microbes at greater depths.

§ 7. *Research for some other specific species of microbes.*

We endeavoured to ascertain the occurrence of obligate-aerobic nitrifying bacteria and of Azotobacter chroococcum, however, with negative result, as could be expected.

Nor could denitrifying microbes be demonstrated; no more could we detect anaerobic butyric bacteria and anaerobic bacteria which break down cellulose.

§ 8. Van der Sleen's *Manganese-Theory for the oxidation of organic matter at greater depths in the soil.*

The problem of oxidation of the organic matter in the deeper layers of the earth has been discussed by W. G. N. van der Sleen

in his publication: „Bijdrage tot de kennis der chemische samenstelling van het duinwater in verband met de geo-mineralogische gesteldheid van den bodem." The writer says (p. 50) that at such a great depth bacterial influence on the oxidation of organic matter seems to be out of the question and he suspects manganese salts to act as oxygen-carriers. Further on (pg. 62) the writer says: „. . . . I do not think that Microspira desulfuricans occurs at such a depth as has to be assumed when ascribing sulphate-reduction only to this micro-organisme".

On pag. 51 the author records some experiments which go to show that manganese can transmit oxygen from the sulphates to an organic compound such as hydrochinon. To conclude from this that the oxidation of organic matter at the lower depths in the soil could occur in the same way, seems to me hardly admissible unless experimental evidence be brought forward that biological oxidation is out of the question. Such evidence has not been produced as yet. It may be deemed surprising that the author, who, as appears from the passage in his publication that we quoted just now, had taken cognizance of the bacteriological sulphate reduction has omitted to inquire into it. This is the more surprising since on the ground of its anaerobic behaviour Microspira desulfuricans is adapted to living at greather depths in the soil.

The evidence produced by our investigation set forth in the preceding paragraphs, by which it has been established that sulphate-reduction is of common occurrence at the greater depths underneath the dunes, warrant the conclusion that oxidation of organic matter can be effected by Microspira desulfuricans, without the additional influence of manganese compounds.

§ 9. *The transformation of sulphate in the clay-containing soil of the dunes and sulphate-reduction by Microspira desulfuricans.*

The "Koninklijke Academie van Wetenschappen te Amsterdam" [1]) has brought forth a report on the question to what the presence of so called Artesian water in the dune-soil is due, in a preliminary advice from G. A. F. Molengraaff and Eug. Dubois. In an enumeration of the chemical properties of dune-water the report contains the following statement:

---

[1]) Verslag v. d. gewone vergaderingen der Wis- en Natuurk. Afd. Vol. XXX, p. 212.

"From the surface downwards in and underneath the dune-masses the sulphuric acid content diminishes proportionally to the total thickness of the clay-layers occurring in them, i. e. in proportion to the increase of the volume of clay-soil, through which the water has percolated downwards.

This phenomenon is the result of the power of clay-soil to convert sulphuric acid and then retain it".

In the study by Eug. Dubois [1]) already quoted above, a detailed exposition is given of the transformation of sulphuric acid in the clay-layers, which consists in a reduction-process in the presence of organic matters, with formation of ferric sulphid.

It is evident from the foregoing that sulphate-reduction, which occurs not only in the deeper clay-layers, but also in the sand-soil, is brought about by Microspira desulfuricans. The life of this microbe, which is adapted to anaerobic conditions, accounts for the common occurrence of sulphate-reduction in the deeper layers of the earth and especially in the clay-soil, which generally has a higher content of organic matter.

So long as the conditions of this typically microbiological process are fulfilled, transformation of sulphate into ferric sulphid will hereby be generated, to which is to be ascribed the partial or total absence of sulphuric-acid salts in deep-dune water.

Heemstede, February 24 1922.

---

[1]) „Het Leidsche Duinwater". Een hydrologische studie, 1912, p. 20.

Chemistry. — "*The Influence of a Catalyst on the Thermodynamic Quantities Regulating the Velocity of a Reaction.*" By E. VAN THIEL. (Communicated by Prof. J. BÖESEKEN.)

(Communicated at the meeting of May 27, 1922).

According to GULDBERG and WAAGE's hypothesis the velocity of reaction in a homogeneous system of constant temperature is equal to the product of the active masses of the converted substances multiplied by the velocity constant. This constant is, of course, variable with the temperature, and dependent on the nature of the reacting molecules. The differential equations of G. and W. only indicate the number of molecules decomposed in the time unit; they do not indicate precisely how the reaction(s) takes place; hence they do not show either how the reaction constant depends on the nature of the substances and on the temperature.

Disregarding NERNST's formulation, in which the velocity of reaction is put $= \frac{\text{chemical force}}{\text{chem. resistance}}$, a formula that proved untenable, GOLDSCHMIDT's attempt [1]) to give an explanation of the nature of the reaction constant may be called the first. Starting-point for these and following theories were especially two considerations referring to bi-molecular gas reactions:

1. the reaction constant (velocity for concentration $= 1$) is doubled about per 10° of increase of temperature, so long as the observations are not too far from room temperature. The number of collisions of the molecules is proportional to the translatory velocity, hence proportional to $\sqrt{T}$. The increase of this kinetic energy can, therefore, contribute to a maximum of 2% to the increase of the velocity of the reaction found. Hence a deeper insight into the nature of the reaction than is given by G. and W.'s theory is necessary.

2. if all the molecules of the decomposed gas were in the same state, every collision would be followed by a reaction. Every reaction would then take place with the same explosive velocity. This not being the case, all molecules are not equally reactive. A fraction of them is in a more favourable condition. It is, therefore, possible that the velocity of reaction is proportional to the number of these

[1]) Diss. Breslau, 1907. Cf. also TODD and OWEN, Phil. Mag. 37, 224.

favoured molecules. Whether it is necessary for the reaction that *two* active molecules collide, or whether it is sufficient when *one* of them is active, must for the present remain an open question.

GOLDSCHMIDT assumed that the velocity of reaction is about proportional to the number of molecules the translatory velocity of which exceeds a definite minimum value. Only *these* molecules, the number of which is given by MAXWELL's law of partition, would be active. This restriction to the velocity of translation, is however, entirely unfounded; it is on the other hand more probable that also the intermolecular and interatomic energies play a part in the reaction, it is, therefore, more plausible to assume a threshold value also for these energies.

KRÜGER's theory is of a more exact character; it has, however, only been elaborated for the simplest cases, as e. g. the dissociation $I_2 \rightleftarrows 2I$, in which the reacting substances are already in atomic (active) condition. TRAUTZ gave a more general theory of velocities of reaction. Starting from VAN 'T HOFF's reaction-isochore:

$\frac{d \ln K}{dT} = -\frac{Q}{RT^2}$, he substituted for $K = \frac{k_1}{k_2}$ and for

$$Q = Q_0 + \Sigma \nu_1 \int_0^T c'_v \, dT - \Sigma \nu_2 \int_0^T c''_v \, dT.$$

He further assumed that k_1 resp. k_2 depends only on the properties of the initial resp. resulting substances, and therefore split the reaction isochore into two parts, each referring to this. For this it must also be possible to split Q_0 rationally, for which purpose T. introduces the conception *intermediate substances* (which have an exceedingly short period of existence). In the case of the splitting up of $2HI \rightleftarrows H_2 + I_2$ these intermediate substances might be H- and I-atoms. For the decomposition of HI into H- and I-atoms a dissociation energy is again required, in the formation of H_2 and I_2 from these atoms a heat Q_2 is liberated. It is clear that $Q_0 = Q_1 - Q_2$. Now all the obstacles to the splitting up of the reaction isochore were removed, and the following equation resulted:

$$\frac{d \ln k_1}{dT} = -\frac{Q_1}{RT^2} - \Sigma \nu_1 \int_0^T c'_v \, dT / RT^2.$$

By integration and further elaboration TRAUTZ obtained a formula which in approximation could be reduced to a considerably simpler form, and from which some important conclusions may be drawn:

$$\log k = (25 \text{ to } 35) - \frac{Q_1}{4{,}571\, T} - \Sigma\, \nu\, .\, 1{,}5 \log T - \Sigma\, \nu\, .\, 1{,}1$$

while the following form may be derived for the temperature coefficient of the reaction constant:

$$\frac{k_{T_2}}{k_{T_1}} = e^{\frac{Q_1}{R} \cdot \frac{T_2 - T_1}{T_2 T_1} - \frac{\Sigma\, \nu}{R} \int \frac{dT}{T^2} \int_0^{T_2} c_v\, dT + \frac{\Sigma\, \nu}{R} \int \frac{dT}{T^2} \int_0^{T_1} c_v\, dT.}$$

From the first of the two equations may be read that in bi-molecular reactions the velocity is *greater*, as the Q_1 is *smaller*, while it follows from the second equation that the temperature coefficient *increases* on *increase* of the Q_1. When, therefore, the same reaction is brought about without, and one with exceedingly little catalyst under for the rest identical circumstances, the catalysed reaction, which proceeds more quickly, will require a smaller heat of activation for its molecules, and possess a smaller coefficient of temperature than the not catalyzed reaction, two conclusions which may be verified experimentally.

Briefly T.'s train of thought comes to this that he assumes that it is required both on formation and decomposition of molecules that they pass into a reactive form (not always atoms) with absorption of energy and that on collision of these active molecules the reaction always takes place. VAN 'T HOFF's reaction isochore being the starting point in TRAUTZ's theory, it is comprehensible that the stress has been laid on the changes of energy taking place in the reaction, and that the importance of the constant of integration is not sufficiently brought out. And yet it is clear on some consideration that the only thing required for a bi-molecular reaction is not a collision, but a collision at the right place (perhaps with the exception of very simple molecules). This favourable constellation which may be expressed in the form of a quantity of entropy, does not occur in the reaction isochore. Accordingly in TRAUTZ's theory changes of entropy in the reaction have only been considered in so far as the number of collisions are concerned.

That with by far the majority of the reactions change of the internal energy of the molecules is accompanied with change of the molecular entropy [1]), is not sufficiently taken into account in LEWIS's theorie either. There, too, it is assumed that before being able to react, every molecule must have a certain excess of energy, called by L *increment of energy*. This increment would be absorbed in

[1]) TRESLING, These Proc. Vol. XXIII, p. 143.

the form of infra-red radiation of very definite frequency, which radiation is present in the medium in virtue of its thermal condition. By the application of PLANCK's law of the normal partition of energy, the density of radiation of this frequency can be calculated at every temperature, and from this the fraction of the molecules that are in reactive condition. LEWIS derives that the increment of energy E. is equal to a quantum (of the absorbable type) per molecule.

$$E = Nh \Sigma \nu_{reagents.}$$

LEWIS derives for the constant of reaction of a bi-molecular reaction:

$$k = P_0 \sqrt{T}\, n_A^3\, n_B^3\, e^{-h(\nu_A + \nu_B)/kT}$$

in which $P_0 =$ constant, $T =$ absolute temp., n_A and $n_B =$ index of refraction of the substance A resp. B, and $k =$ gas constant per molecule. The formula shows very clearly the rapid increase of k on rise of temperature.

The nearer the value of the critical energy is to that of the mean energy per molecule, the greater will be the number of molecules becoming active per second, hence also the velocity of reaction, the same conclusion, therefore, to which TRAUTZ came. By taking the logarithm of the above formula, and differentiating this with respect to time, the following form results:

$$\frac{d \ln k}{dt} = \frac{Nh(\nu_A + \nu_B) + {}^1/_2 RT}{RT^2} = \frac{E + {}^1/_2 RT}{RT^2}$$

Of reactions which take place as much as possible under the same circumstances, only more slowly or more quickly (to be realized with little catalyst), the quicker reaction must have a smaller E according to the above, from which it then follows that $\frac{d \ln k}{dt}$ has become smaller, hence also the temperature coefficient of the reaction constant, for the temp.coëf.

$$\frac{k_{T+10}}{k_T} = \frac{k_{T+10} \frac{dk}{dt}}{k_T} = 1 + 10 \frac{dk}{k_T\, dt} = 1 + 10 \frac{d \ln k}{dt}.$$

LEWIS (like TRAUTZ) draws the conclusion that a strongly catalyzed reaction will indicate a decrease of the temperature coefficient compared with the same reaction weakly or not catalyzed.

On half thermodynamic, half kinetic grounds KOHNSTAMM and SCHEFFER have derived a relation between the velocities of reaction and the thermodynamic potentials of the substances participating in

a reaction. Starting from this SCHEFFER drew up a simple formula which agrees with a formula derived at about the same time in an entirely different way by MARCELIN, viz.:

$$ln\, k = -\frac{E}{RT} + B,$$

in which E represents the difference of energy between the intermediate state which is rich in energy, and the mean condition of the reacting substances in the reaction, and B is a quantity which does not contain constants dependent on the nature of the substances, except the difference of entropy. This term takes the effective chance of collision into account. It follows from the formula that increase of the energy increment diminishes the velocity of reaction, increase of the difference of entropy on the other hand increases it. In contradistinction with the formulae discussed before, a catalyst need not necessarily decrease the energy increment; it is even possible that as a result the energy increment is increased, provided the increase of factor B more than neutralizes this unfavourable action. The increase of the energy increment means fewer active molecules, increase of B is equivalent to a more favourable chance of collision. It is, therefore, possible that the action of a catalyst would consist in this that though the threshold of energy should be raised, the number of favourable collisions has been so much increased that the reaction nevertheless proceeds more rapidly.

In the not catalysed reaction by no means every collision between active molecules would eventuate in a reaction. This is a priori sooner to be expected for complicated than for simple molecules; instances are, therefore, especially to be found in organic chemistry.

From increase of the energy-increment ensues increase of the temperature coefficient, hence the catalysed reaction can have a greater temperature coefficient than the not catalysed reaction. *Entirely in contradiction with* TRAUTZ's *and* LEWIS's *conclusions the catalysed reaction can have a temp. coef. and an energy increment which are greater than those of the same reaction without catalyst.*

Measurements of the velocity of one and the same reaction between complicated molecules with and without catalyst and at different temperatures might give a decision in favour of SCHEFFER's theory, if a reaction could be found which, catalysed, presented a greater temperature coefficient than not catalysed. As will be seen in what follows, this appeared to be the case in the acetylation of diphenylamin.

The reaction was carried out at 0°, 20°, 30°, 40°, and 50° C.

The excess of acetic acid anhydride was taken so great that the variations of concentration of this component could be neglected with respect to those of the component diphenylamin. Hence the reaction was pseudo-mono-molecular. Many catalysts were tried [1]) before some substances were found which were not paralysed during the reaction; they were p. bromo-benzene-sulphonic acid and p. toluene-sulphonic acid.

The following tables give the observations from 0—50° without catalyst.

temp. 0° 1 mol diph. 12½ mol. anh.

t	% converted	$\frac{K}{2.303}$
1.— uur	1.1	0.0048
2.— „	1.4	0.0031
3.— „	2.1	0.0031
4.— „	2.3	0.0025
5.— „	2.4	0.0021
6.15 „	3.0	0.0021

temp. 20° 1 mol. diph. 12½ mol anh.

t	% converted	$\frac{K}{2.303}$
0.30 uur	0.8	0.0069
1.30 „	2.4	0 0070
2.30 „	3.8	0.0067
3.30 „	5.5	0.0070
4.30 „	7.3	0.0073
6.— „	9.1	0.0069

temp. 30° 1 mol diph. 12½ mol. anh.

t	% converted	$\frac{K}{2.303}$
0.30 uur	1.6	0.0127
1.01 „	2.8	0.0121
2.— „	5.5	0.0123
3.— „	8.2	0.0124
4.— „	10.8	0.0124
6.05 „	16.3	0.0129

temp. 40° 1 mol. diph. 12½ mol anh.

t	% converted	$\frac{K}{2.303}$
0.33 uur	2.5	0.0220
0.58 „	4.3	0.0208
2.— „	8.9	0.0202
2.59 „	12.9	0.0201
5.— „	21.5	0.0210
7.— „	29.0	0.0212

Taking into consideration that in the first table the converted quantities are so small, the most probable values of the reaction constants are respectively; 0,0021—0,0070—0,0124—0,0209 and 0,0384.

---

[1]) Diss. Delft 1922.

temp. 50° 1 mol. diph. 12½ mol. anh.

t	% converted	$\frac{K}{2.303}$
0.30 uur	4.4	0.0391
1.— „	8.4	0.0381
2.— „	15.2	0.0358
3.— „	22.6	0.0371
5.— „	36.1	0.0389
7.— „	46.5	0.0388

The reaction constants of the catalysed reactions are recorded in the following table:

	p.bromo-benzene-sulphonic ac. cat. 0.00089 mol.	0.00178 mol.	p. toluene-sulphonic acid catal 0.00089 mol.	0.00178 mol.
k_0	0.0018	0.0027	0.0021	0.0024
k_{20}	0.0102	0.0197	0.0105	0.0134
k_{30}	0.0243	0.0523	0.0235	0.0340
k_{40}	0.0598	0.143	0.0558	0.0819
k_{50}	0.153	0.383	0.123	0.194

It is remarkable that the activity of the catalyst decreases at low temperatures, and becomes about 0 at 0°. At lower temperature the catalyst is paralysed, to which we shall revert later on. The energy increment can be calculated from two observations by the aid of the formula $ln \frac{k}{k_1} = \frac{E}{R}\left(\frac{1}{T_1} - \frac{1}{T}\right)$. In the calculation of the energy-increment of the catalysed reactions it should be borne in mind that this must not be done in the usual way, if the measured reaction is a combination of two or more reactions taking place side by side [1]). The hypothesis according to which it is assumed that with a small catalyst concentration, the number of collisions of the kind as occur in the non-catalysed reaction, remains the same, and that only another kind of collisions is added to them, is permissible in my opinion. In this case the measured constant of reaction

[1]) Lacomblé, Diss. Leiden 1920, p. 80.

represents the sum of that of the non-catalysed and that of the purely catalysed reaction. In order to obtain the constants of the purely catalysed reactions, which are recorded in the above table, the reaction constants of the non-catalysed reactions must be subtracted from the measured ones. It has been tacitly assumed, what is, indeed, shown by the constancy of the measured reaction constants, that the two reactions proceeding side by side, are of the same order, as otherwise this operation is not allowed.

	p. bromo benzene sulph. ac. catal.		p. toluene sulphonic catal.	
	0.00089 mol.	0.00178 mol.	0.00089 mol.	0.00178 mol.
k_0	—	0.0006	—	0.0003
k_{20}	0.0032	0.0127	0.0035	0.0064
k_{30}	0.0119	0.0399	0.0111	0.0216
k_{40}	0.0389	0.122	0.0349	0.0610
k_{50}	0 155	0.345	0.0846	0.156

The energy increment calculated from the 1st series of observations without catalyst, and from these 2nd, 3rd, 4th, and 5th series is respectively ± 10.000 calories — 23000 cal. — 20500 cal. — 20500 cal. — 20800 cal.

The acetylation of diphenylamin decides, therefore, in favour of SCHEFFER's theory, as it would e.g. be entirely inexplicable according to LEWIS, why the sulphonic acid can act as catalyst, as the addition of this substance about doubles the energy-increment; the number of active molecules would, accordingly, be much smaller, hence also the number of effective collisions.

In the calculation of the factor *B* from SCHEFFER's formula, it appears to be more than doubled by the catalyst. The favourable chance of collision has, therefere, been enlarged, notwithstanding the number of active molecules has become smaller. Hence if the conversion is to be increased, this smaller number of active molecules must collide more favourably. Accordingly every collision between molecules that are sufficiently rich in energy does not always eventuate in conversion, it is probably only a small percentage of them that enters into reaction.

One can form the following conception of this.

It is not immaterial what part of the acetic acid anhydride molecule impinges with the diphenylamin-molecule, nor with what part of the latter. The reactive molecule parts, in this case the

1st series.

	E	B
$k_{20} : k_0$	9600	12.3
$k_{30} : k_0$	9800	12.7
$k_{40} : k_0$	9800	12.6
$k_{50} : k_0$	10200	13.4

2nd series.

	E	B
$k_{30} : k_{20}$	23200	34.7
$k_{40} : k_{30}$	23000	34 4
$k_{50} : k_{40}$	22600	33.8

3rd series.

	E	B
$k_{30} : k_{20}$	20200	31.1
$k_{40} : k_{30}$	20800	32.0
$k_{50} : k_{40}$	20800	32.1

4th series.

	E	B
$k_{30} : k_{20}$	20400	30.1
$k_{40} : k_{30}$	21000	31.1
$k_{50} : k_{40}$	20200	29.8

5th series.

	E	B
$k_{30} : k_{20}$	21600	32.7
$k_{40} : k_{30}$	20800	31.3
$k_{50} : k_{40}$	20200	30.4

oxygen bridge of the anhydride and the aminohydrogen of the amin, must be in each other's immediate neighbourhood. In a substance which exercises an attraction on these two parts, these molecule parts will be turned towards each other at a collision of the three molecules (more probable is a collision of a molecule with the complex of the two others). The sulphonic acids used certainly exert an attraction on the amino hydrogen, and most likely also on the bridge oxygen, because sulphuric acid impinges with the anhydride at that place, and the sulphon group is the active component in both substances. In my opinion the catalytic action of sulphonic acid is for the greater part due to its *directive* action, and it owes this directive action to its affinity towards the reaction components, as BÖESEKEN's dislocation theory demands for every catalyst, without this affinity leading to such a firm bond, that the affinity, hence also the directive action on the other kind of molecule, would be eliminated.

Against these conclusions the question might be raised whether the measured temperature coefficient represents indeed the real one. The nature of the catalyst leads to the supposition that a part of the sulphonic acid is bound to the diphenylamin resp. anhydride (or both), and that this might not be active (or much less so). On rise of temperature a stronger dissociation would appear in the components, hence more free (i.e. more active) catalyst would be present. Then

the measured temp. coeff. is not $= \frac{k_{T+10}}{k_T}$, but $= \frac{k_{T+10}}{k_T} \times \frac{c_{cat\ T+10}}{c_{cat\ T}}$. Accordingly the real temp. coef. would be smaller than the measured one.

Let us suppose the bound catalyst to be totally inactive, and the true temp. coef. to have remained of the same value as was found in the non-catalysed reaction. The measured temp. coëf. $= 3 = 1{,}8 \times \frac{c'_{cat}}{c_{cat}}$, from which $\frac{c'_{cat}}{c_{cat}} = 1{,}67$, in other words one is confronted by the question whether it is possible that in the neighbourhood of room temperature the concentration of the components increases by 67 % per 10° increase of temperature.

Let us take 300° and 310° absolute for the two temperatures, 0,1111 as constant of equilibrium at 300° (hence 90 % bound cat, 10 % free cat), and let us put the heat of dissociation $= 5000$ cal, a heat which may be called normal.

$$K_{300} = \frac{10 \times c_A}{90} \qquad K_{310} = \frac{x \times c'_A}{100 - x}$$

in which $c_A = c'_A$ may be put:

$$\frac{K_{310}}{K_{300}} = \frac{x}{100 - x}\, 9.$$

It follows from the reaction-isochore that:

$$log \frac{K_{310}}{K_{300}} = \frac{Q}{4{,}571} \cdot \frac{T_{310} - T_{300}}{T_{310}\, T_{300}} = \frac{5000}{4{,}571} \cdot \frac{10}{93000} = 0{,}1176$$

from which:

$$\frac{K_{310}}{K_{300}} = 1{,}31 = \frac{x}{100 - x}\, 9$$

from which:

$$x = 12{,}7 \quad \text{and} \quad \frac{c'\ cat}{c\ cat} = 1{,}27.$$

Hence even in the most unfavourable case conceivable that the bound catalyst would be totally inactive, the increasing dissociation per 10° increase of temperature is only able to account for a small part of the increase of the temp. coef. of the catalysed reaction above that of the non-catalysed reaction.

Scheffer pointed out that in many cases the E may be put practically constant over a limited range of temperature, and that in this case B is also pretty well constant. If the region from 20°—50° lies within this limited range, the values of $ln\, k$ drawn

as function of $\frac{1}{T}$, must lie on a straight line, for every equation of the form $y = mx + b$ represents a straight line. Expressed in a graphical representation this appears really to be the case [1]), and the course of the lines suggests that the energy-increment is little, if at all, dependent on the quantity of added catalyst. The values of $ln\, k$ at 0° fall outside the straight line in the catalysed reactions.

As on account of the slight velocity of the reaction at 0° the observations need not be very accurate, I repeated two measurements at 0°, viz. of the non-catalysed reaction, and of that with 0,00178 mol. p. bromobenzene sulphonic acid. I extended the observations over fully two days instead of over seven hours.

temp. 0° 1 mol. diph. 12½ mol. anh.

t	% converted	$\frac{K}{2.303}$
21.55 uur	8.0	0.00165
28.26 »	10.1	0.00162
44.55 »	14.5	0.00152
52.52 »	17.6	0.00159

temp. 0° 1 mol. diph. 12½ mol. anh. 0.00178 mol. acid.

t	% converted	$\frac{K}{2.303}$
21 48 uur	10.3	0.00216
28.19 »	12.9	0.00212
44.48 »	19.4	0.00210
52.44 »	23.8	0.00224
75.46 »	32.3	0.00226

Though the values which I found, were indeed lower, the constant of the purely catalysed reaction appeared to have the same value as was determined in former experiments, viz. 0,00218—0,00160 = = 0,00058 (found formerly = 0,0006).

To be expected was a value $\pm \frac{0{,}0127}{(3{,}1)^2} = \pm$ 0,00132, hence for the gross catalysed reaction 0,00132 + 0,00160 = ± 0,0029; a value that exceeds the error of observation many times.

In $FeCl_3$ I think I have found a catalyst which is catalytically active undiminished down to 0°. As these experiments have not yet been completed, they will be discussed in a later publication; I may conclude from the experiments already made that also ferri-chloride enlarges the "hill" of energy and that accordingly also this catalytic action can alone be explained by the aid of SCHEFFER's theory.

[1]) Diss. Delft 1922.

Chemistry. — "*The Dislocation Theory of Catalysis.*" By Prof. J. Böeseken.

(Communicated at the meeting of May 27, 1922).

The explanation of the catalytic phenomena has always presented great difficulties, and has never been satisfactory as yet, because the cause of changes of reaction-velocity was to be ascertained without there seeming to be a clear relation between the velocity of reaction and the quantity of energy that came into play.

Before the catalytic phenomena had been brought in connection with the conception of free energy, satisfaction might be found in establishing the fact that one or more intermediate reactions took place, which together proceeded more rapidly than the reaction without catalyst.

And it is still possible to be satisfied with such an explanation when it can also be shown that the catalyst in quantity and quality is eventually regained unchanged from the reaction mass.

It should, however, be fully realized that no answer is given to the question why these intermediate reactions proceed more rapidly than the principal reaction.

This is the more striking, because in these intermediate reactions the catalyst disappears from the reaction-mass at least temporarily and partially. I have, therefore, pointed out that the ideal catalysts are exactly those that are *not* fixed in intermediate reactions, and that the *real catalysis* is the interaction between the catalyst and the molecules, which has nothing to do with the formation of a compound as such.

This interaction, which I have called *dislocation*, may be seen as a change of the paths of the electrons; it is very well possible that it cannot take place until the catalyst has formed a compound with the molecules, but at any rate it must be possible to show it in some way or other and to express it in a mathematical form.

On one side it is therefore necessary to form a clear conception of the dislocation, on the other side the modifications which take place in the thermodynamic relations through the presence of a catalyst and to which the changes of the reaction velocities respond, must be fixed in a mathematical formula by establishing a connec-

tion between the reaction velocities and the thermodynamic relations.

As regards the former, *in the oxidation of alcohols with cooperation of aromatic ketones activated by light* I have found a reaction, in which the catalysis proper (the dislocation) could be sharply distinguished from the formation of a coumpound between catalyst and the molecules present.[1])

When an alcoholic solution of benzophenon, which is kept saturate with oxygen, is exposed to violet light, the alcohol is oxidized to aldehyde and water, the ketone remaining unchanged.

A closer study brought to light that above a certain concentration of the ketone the velocity of reaction became independent of this concentration, and further that it was proportional to the *square* of the intensity of the light and to the first power of the conc. of the alcohol.

This may be explained as follows:

The ketone absorbs part of the light and is activated by it. According to the laws of absorption the quantity of active ketone will be proportional to:

$$I(1-e^{-kcd}) \text{ in which } \begin{array}{l} k = \text{absorption coefficient} \\ c = \text{concentration ketone} \\ d = \text{thickness of layer} \end{array}$$

Ie^{-kcd} is the light that is transmitted. If k, c, and d are pretty great, this is very little, and all the light is absorbed. The quantity of activated ketone then becomes proportional to I and independent of c, its concentration.

When we assume that among others the two following processes take place:

$$2 \text{ active ketone} + \text{alcohol} = (\text{active ketone})_2 \text{ alcohol}$$

and

$$(\text{active ketone})_2 \text{ alcohol} + O = \text{ketone} + \text{aldehyde} + H_2O,$$

the former of which proceeds much more slowly than the latter, the oxygen absorption (which was measured) will be determined by the first process, the velocity of formation of the ternary compound. This velocity of reaction must then be proportional to the square of I and to the concentration of the alcohol.

I will not enter here into a fuller discussion how this might be proved in different other ways[2]). The whole process can now be described as follows:

Under the influence of the light there is suddenly formed a

[1]) Recueil **40**, 433—445.

[2]) l. c. p. 439—442.

quantity of photo-ketone $= kI\ (1 - e^{-kat})$ approaching to kI.

I. ketone + light = photo-ketone;

as this quantity is formed at the moment that the solution is illuminated, it is as if with the velocity of light a plate of a catalyst slides on the light side of some vessel or other, in which the solution is put.

Then the reaction takes place the velocity of which regulates the process: the meeting of the alcohol molecules and those of the photo-ketone:

II. 2 photo-ketone + alcohol = (photo-ketone), alcohol.

By this meeting two H-atoms of the alcohol are activated:

III. (photo-ketone), alcohol → [(photo-ketone), active alcohol].

This process, which probably takes place with the velocity of light, *is the real catalysis, the dislocation.*

The alcohol-molecules are enabled to react with the oxygen according to the scheme:

IV. 2 [(photo-ketone) active alcohol] + O_2 =

4 ketone + 2 aldehyde + $2H_2O$,

which last process also takes place with great velocity.

We see that the actual catalysis has to do with the formation of the ternary compound only in so far as the photo-activity of the C = O-groups of the ketone can be transferred to the H-atoms of the alcohol. Here the distinction of the catalysis and of the formation of the compound is, indeed, very clear, for the ternary compound is also formed in the dark, and then there is no question of any catalytic action.

When the photo-ketone is thought replaced by an ordinary catalyst, e. g. a plate of paladium, it is clear that the combining of this metal with the alcohol is not the essential part of the catalysis, but what happens with the alcohol molecules at the moment that the atoms *Pd* get into contact with them, through which two of the H-atoms are activated. This the paladium can do by itself, without being activated by a stimulus from outside.

It appeared from the light-investigation that the oxidation of the activated alcohol molecules took place very rapidly. This will as a rule also be the case in the ordinary catalysis, but this velocity can be different for each case.

If, however, a catalyst in very small quantities is to accelerate a given reaction considerably, every contact of its molecules with those of the substance that is to be activated, must give rise to a dislocation that sets in very rapidly.

It is clear that this can hardly take place otherwise than on *intimate* contact, and here the significance of the formation of a compound between catalyst and the molecules to be activated, even though it be one that can very easily be dissociated, comes to the fore. In the light investigation it was only the primary and secondary alcohols that were easily oxidized, and *not* the hydrogen itself and a number of hydrogen compounds, evidently because the former *could*, the latter could *not* be attached by the ketone.

As has been said in the introduction, not only must the conception of dislocation be defined more closely, but it must also be tried to find a mathematical form for it through the consideration of the thermo-dynamic and kinetic relations.

Of late years many scientists have occupied themselves with studies of the reaction velocities, which are also the subject of this investigation. We may mention the names of TRAUTZ, MARCELIN, LEWIS, PERRIN, and SCHEFFER [1]).

It seems to me that SCHEFFER's considerations have the greatest value for the knowledge of the significance of the *dislocation*, because there the question is put whether a formula for the phenomena of diffusion (drawn up by KOHNSTAMM) is also valid for the description of the reaction velocities, and this question is answered in an affirmative sense.

For the chemical phenomena are essentially phenomena of diffusion in which particularities will occur only in the partial mutual penetration of atoms and molecules. In SCHEFFER's theoretical research the significance of these particularities, which were represented in the form of thermo-dynamic relations of the "intermediate states", was clear. It is self-evident that it is exactly *these* relations which are modified by the catalyst, and that comparison of these relations without and with the catalyst, must lead to a standard of the *dislocation*.

SCHEFFER's simple equation of the relation between the reaction constant, the quantities in question, and the temperature is:

$$ln\, k = -\frac{E_t - E}{RT} + B.$$

In this $E_t - E$ is a measure for the difference of energy between the reacting substances and the intermediate state at the reaction [2]), B contains the differences of entropy and constants

[1]) These Proceedings Vol. XIII, p. 789 and Vol. XV, p. 1109.

[2]) It is the energy which a gram-molecule requires above the mean energy at the temp. T in order to react, and which is sometimes expressed by the name of *energy-increment*.

which do not depend on the nature of the reacting substances.

As a difference of entropy is a measure for a greater or less probability, and as this probability must refer to the reaction setting in more or less easily, both this difference of energy and this probability can be calculated by the aid of this formula from two observations at different temperatures, and by carrying out this calculation with and without a catalyst it can be ascertained in what way these two relations are modified by the catalyst.

As appears from the following communication, this calculation has been applied by E. VAN THIEL to the acetylation of diphenylamin with acetic acid anhydride both in presence of *p*-bromo (methyl)-phenylsulphonic acid as catalyst and without it, and the remarkable result has been obtained that in the presence of a catalyst the factor $(E_t - E)$ is about doubled, B becoming also considerably larger. The conclusion may be drawn from this that in this case by the addition of a catalyst more than double the energy is, indeed, required to cause the molecules to react than without it, but that this unfavourable factor is far more than compensated by the so much greater probability for the setting in of the reaction in the presence of the catalyst.

In his address at the spring meeting of the Ned. Chem. Ver. (Dutch Chemical Association) of April 20 1922 SCHEFFER expressed this as follows: the hill of energy that is to be surmounted becomes, indeed, higher, but the road across it, becomes very much broader.

Though it may be more or less a coincidence that in the case examined by VAN THIEL the hill of energy is so much higher in the presence of a catalyst than without it, it is yet the confirmation of my view that the formation of a compound between the catalyst and the substances to be activated sooner hampers than promotes the reaction, and that the catalyst performs its accelerating action *not by combining* with these molecules, but *in spite* of this combination.

The acceleration of the reaction takes place because simultaneously with the formation of this compound a change of condition sets in, the *dislocation*, in which the conditions for the occurrence of the reaction become so much more favourable. The conception of dislocation has found a confirmation through SCHEFFER's theoretical investigation, and a measure in the variation of the quantity B of his formula.

In conclusion it may still be pointed out that the thermo-dynamic-kinetic considerations have not brought the question *why* a catalyst creates favourable conditions, nearer to its solution. The possibility may be considered of the molecules assuming a certain position,

which causes the collisions to take place on the reactive parts of the molecules (VAN THIEL, see following communication), or it may be supposed that the reactive surface is enlarged, etc. etc. It is certain that these changes of position or of form must take place very rapidly, and the catalyst must be under very favourable conditions with regard to the molecules that are to be activated, which can hardly be imagined in another way than ensuing from a chemical affinity, which leads to dissociation equilibria that are established very rapidly.

Delft, May 1922.

Botany. — "*The disappearance of the diploid and triploid magnicoronate narcissi from the larger cultures and the appearance in their place of tetraploid forms*". By Dr. W. E. DE MOL. (Communicated by Prof. G. VAN ITERSON JR.).

(Communicated at the meeting of June 24, 1922).

I. *Introduction.*

Simultaneously with my investigations into the causes which lead to the immense variety of size and form in the *Hyacinthus orientalis* in Holland, I commenced a similar research with respect to the species of narcissi and narcissus-hybrids under cultivation. These comparative researches have led to some noteworthy results. One conclusion I arrived at was that, as is the case with *Hyacinthus orientalis*, the remarkable size of the bulbs, leaves and flowers which characterize the bastards of *Narcissus Pseudonarcissus* now cultivated, correspond mainly with the number of chromosomes of which, according to my cytological observations, the somatic nuclei consist.

This feature which, as far as I have been able to observe, occurs in *Hyacinthus orientalis* only in the Dutch cultures, is found both in England and in the Netherlands in *Narcissus Pseudonarcissus*, and is more pronounced than in the hyacinth. In the latter there are probably no tetraploid plants yet, whereas there are several in the *Narcissus Pseudonarcissus*.

II. *Some results of the cytological investigation.*

The preparations which I used in my cytological researches were made in the same way as those for hyacinths. The thickness of the sections is 10 or 15 μ according to the size of the cells and nuclei.

ASCHERSON and GRAEBNER (1) give the *Magnicoronati* as the 1st. section of the sub-genus *Eunarcissus*. This section is entirely formed by the class *N. Pseudonarcissus*, which they divide into 2 subclasses, *N. festalis* and *N. minor*. For convenience sake in describing the varieties studied, I shall keep to this classification, except that I shall place the sub-division *N. minor* first.

1. *N. minor.*

The somatic nuclei of *N. minor* (the type), *N. nanus*, *N. minimus* and *N. cyclamineus* (which is best classed with the sub-species

N. minor) consist of 14 cylindrical chromosomes, 10 long ones and 10 short ones.

2. *N. festalis.*

a. Diploid varieties.

The somatic nuclei of *N. muticus* (syn. *abscissus*), *Capax plenus* (which perhaps ought to be classed under *N. minor*), *Telamonius plenus* (Double Sion, Wilmer's great double golden yellow Daffodil), large old double yellow trumpet Daffodil) also comprise 14 chromosomes which I cannot distinguish from the former ones.

b. Heteroploid varieties.

N. Johnstoni Queen of Spain possesses somatic nuclei with 20 chromosomes. In *Maximus* and *Golden Spur* these nuclei consist of 21 chromosomes, so that judging from the number these varieties are triploid.

The nuclei of *Bicolor Victoria* and *Buttonhole* (obtained from *Bicolor Victoria* by budvariation) contain 22 chromosomes. The chromosomes-garniture of both forms is the same.

The varieties *King Alfred* and *van Waveren's Giant* are, to judge from the number of chromosomes, tetraploid, for here the somatic nuclei consist of 28 chromosomes.

In all the 14 forms above-mentioned and examined, the chromosomes — both long ones and short ones — correspond in size and shape. The diploid nuclei always consist of 10 longer and 4 shorter chromosomes. I cannot yet state the exact number of long and short chromosomes of the nuclei of the heteroploid forms. To do this it is necessary to examine over 3000 good sections with dividing nuclei; I have now examined this number. Probably the longer and shorter chromosomes do not differ in length and breadth from each other, and as in *Hyacinthus orientalis* the pairs of long and short chromosomes will not be distinguishable from each other by any characteristic constant difference in form, as is described of *N. poeticus* by Stomps (3).

III. *Self-pollination in diploid, triploid and tetraploid forms.*

In contrast with *Hyacinthus orientalis*, in such categories as can be distinguished cytologically, self-pollination yields good practical results. From the few seeds of the diploid *N. minimus*, *minor*, *cyclamineus* (and *N. triandrus albus*), taken in 1913, 1914 and 1915, I have reared plants which are not distinguishable in bulb leaf and flower from the parent species.

In the case of the triploid *Golden Spur* self-pollination yielded

plants which in form and size differed from each other and from the parent species.

By means of self-pollination of the tetraploid *King Alfred* I got hundreds of seeds in 1914 and 1915. In 1916 I had about 1400 small bulbs. This spring 50 flowers came out, which differed greatly in form and size from each other and from *King Alfred*. Most of them were smaller than the parent species. The tetraploid *Van Waveren's Giant* can also be self-pollinated successfully.

IV. *Conclusion.*

1. Of the variety *Maximus* which I examined we are aware that it was already known in 1600, from which it may be inferred that even three hundred years ago there was triploidia in the magnicoronate narcissi. Triploidia must have commenced with the wild species or those again run wild, as the above-mentioned variety and *Golden Spur* (first cultivated between 1885 and 1888) were probably not obtained in nurseries (see 6). Regarding the wild variety of *N. Johnstoni Queen of Spain*, BAKER assumes that this is a hybrid between *N. Pseudonarcissus* and *N. triandrus*. If this is correct — and the bastards cultivated of these two varieties leave no room for doubt — this variety of *Queen of Spain* is in all probability a bastard between a heteroploid form of *N. Pseudonarcissus* and *N. triandrus*, as my experience shows the latter to be diploid and to possess the same chromosome garniture as the diploid narcissi already mentioned.

2. If we keep to the classification of ASCHERSON and GRAEBNER we shall see that the feature of the heteroploidia was first seen in the genus or group of *N. Pseudonarcissus festalis major*, the diversity which by hybridization has principally yielded the large garden forms of the present day.

It is very interesting how the increase in the size of these varieties now cultivated can be traced. Up till 1885 — the diploid varieties were chiefly grown. The culture of the *Golden Spur* marks the beginning of the era of the triploid garden forms.

Bastards between *Maximus*, *Golden Spur* and other valuable kinds are grown, with the result that larger specimens have been obtained, of which *King Alfred* (England; firm of KENDALL) is the finest. From this dates the advent of the tetraploid varieties (1899).

Just as the climax in point of size of the diploids seems to have been reached in *Telamonius plenus*, and of the triploids in *Golden Spur*, the culminating point among the tetraploid forms seems to have been reached in *Van Waveren's Giant*. Nevertheless this

has been surpassed again by magnicoronate narcissi, the dimensions of which are greater in one or two respects (e. g. *Early Giant*, *Apotheose*, *Ajax Grand Vizier*, *Imperator* and *Mammoth*; (see for this the "Weekblad voor Bloembollencultuur", 32nd. Year, 1922, Nos. 85, 87, 89, 91 and 93), so that we may suppose that there are already hypertetraploid forms. In this connection the significant question arises as to whether the number of chromosomes may go on increasing indefinitely. Or, in other words: Is there any limit, and if so, where?

The same question has been asked by Beumer with regard to the increasing size. ("Weekblad" n°. 101). In the following table some of the measurements are given in millimetres; they are nearly the same as those given in the publication of Sydenham (4), with the exception of those for *Mammoth*, which are mentioned in "Weekblad", n°. 93.

Name of variety	Diameter perianth	Tepals		Paracorolla		Number of chromosomes
		length	breadth	length	breadth	
Queen of Spain	82	35	15	28	28	20
Bicolor Victoria	101	44	35	44	44	22
King Alfred	107	40	28	44	50	28
Van Waveren's Giant	127	50	38	50	50	28
Mammoth	140	?	?	55	60	?

3. It goes without saying that I cannot now sacrifice the plants that I have obtained from *King Alfred* and *Golden Spur* for a cytological examination. But even without this examination it seems to me highly probable, especially when I test these observations by those conducted by Winkler with *Solanum* (5) and those of van Overeem with *Oenothera* (2), that these conspicuous differences in form and size are primarily due to an unequal distribution of the chromosomes in the reduction-dividing of which an unequal combination of the sex nuclei is the inevitable result.

LITERATURE.

1. Paul Ascherson and Paul Graebner. Synopsis der mitteleuropaïschen Flora, Bd. 3, Leipzig, Wilhelm Engelmann, 1905—1907.

2. Casper van Overeem. Ueber Formen mit abweichender Chromosomenzahl bei *Oenothera*. Beihefte zum Bot. Centralbl., Bd. 38, Abt. I, Heft 2, 1921.

3. Theo J. Stomps. Gigas-Mutation mit und ohne Verdoppelung der Chromosomenzahl. Zeitschr. f. ind. Abst. u. Vererb. Bd. 21, Heft 2, 1919.

4. Robert Sydenham. All about *Daffodils*. Sec. edition, Midland Daffodil Society, 1911.

5. Hans Winkler. Ueber die Entstehung von genotypischer Verschiedenheit innerhalb einer reinen Linie. Deutsche Gesellschaft für Vererbungswissenschaft. Bericht über die Gründung und die erste Jahresversammlung. Leipzig. Borntraeger, 1921.

6. D. M. Wüstenhoff and R. H. Beerhorst. De Narcis. Leyden, Batteljee en Terpstra, 1908.

Plant physiological Laboratory of Prof. Ed. Verschaffelt,
Hortus Botanicus, at Amsterdam.

Mathematics. — *"Numbers of Circles Touching Plane Curves Defined by Representation on Point Space."* By L. J. SMID Jr. (Communicated by Prof. HENDRIK DE VRIES),

(Communicated at the meeting of June 24, 1922).

The circles of a plane (degenerations included) may be represented without exception through a one-one representation on the points of a projective space. (R. MEHMKE, Zeitschrift für Mathematik und Physik 24 (1879)). We can arrive at it among others in the following way:

Let W be an umbilical point of a quadric O^2 and let w be the tangent plane at that point, B a plane parallel to w. A plane section of O^2 with its pole relative to O^2 is projected out of W on B as a circle with its centre, and inversely. We consider this pole as the image of the circle.

As a special case we may take for O^2 a quadric of revolution of which W is a vertex. If moreover O^2 is a sphere, we get the representation of Prof. JAN DE VRIES (Verhandelingen 29); if W moves to infinity it becomes the representation of Dr. K. W. WALSTRA (Verhandelingen 25).

Prof. HK. DE VRIES has studied cyclographically the circles touching a curve C in B of the order μ, the class ν, passing ε times through both the circle points (with ε different tangents in finite space which cut C at those points in $\varepsilon + 1$ points), touching the line g_∞ singly in σ different points and having further no other singularities than δ nodes, $\varkappa$ cusps, τ bi-tangents and ι inflexional tangents (Verhandelingen 8).

We arrive at the same results through the above mentioned representation. We shall only consider the principal ones.

The curve C is projected out of W on O^2 as a curve consisting of the two generatrices through W, counted ε times, and a curve k of the order $n = 2\mu - 2\varepsilon$ passing $(\mu - 2\varepsilon)$-times through W. σ pairs of tangents at W coincide, because the parabolic branches of C give rise to cusps of k in W. Further k has δ nodes, $\varkappa$ cusps and $(\mu - \varepsilon)$ $(\mu - \varepsilon - 1)$ apparent nodes and no stationary tangents. By means of PLÜCKER's formulas we find other numbers characteristic of k.

From the nature of the representation there follows that the points of the surface L of the tangents of k represent the circles cutting C at right angles. The tangent planes to O^2 at the points of k

envelop a developable surface K the points of which represent the circles touching C and the points of the cuspidal curve l of K represent the osculation circles of C. There exists a polar relation between the points, tangents, and planes of osculation of k and the planes of osculation, tangents, and points of l. Out of the characteristic numbers of k and L we find accordingly through dualisation the characteristic numbers of l and K, for instance:

Order of l: $m = \iota + 3\mu - 6\varepsilon - 2\sigma$

Order of K: $r = 2\mu + \nu - 4\varepsilon - \sigma$

Cusps of l: $\beta = 5\mu - 3\nu + 3\iota - 8\varepsilon - 3\sigma$

Order of the nodal curve of K: $x = \frac{1}{2}\{(2\mu + \nu - 4\varepsilon - \sigma)^2 - 13\mu - \nu - 3\iota + 24\varepsilon + 7\sigma\}$.

From this follows among others:

To a given pencil there belong r tangent circles of C, but to a concentric pencil only $r - (\mu - 2\varepsilon)$ in finite space (class of the evolute). If we have 3 curves C_1, C_2, C_3, the surfaces K_1, K_2, K_3, have in all $r_1 r_2 r_3$ points in common, of which however there lie $4(\mu_1 - 2\varepsilon_1)(\mu_2 - 2\varepsilon_2)(\mu_3 - 2\varepsilon_3)$ in W. The rest is the number of circles touching the 3 curves.

Through a given point there pass m osculating circles of C. The projection of l out of W on B is the evolute; l passes σ times through W, hence the order of the evolute is $m - \sigma$. The evolute has β cusps (vertices of C) in finite space and moreover $\mu - 2\varepsilon - 2\sigma$ at infinity, arising because $\mu - 2\varepsilon - 2\sigma$ tangents of l pass through W, lie in w and have their points of contact outside W.

Through a point there pass x circles touching C twice. The locus of the centres of these bi-tangent circles is the projection of the nodal curve of K. This curve however passes $s = (\mu - 2\varepsilon)(\mu - 2\varepsilon - 1) - \sigma$ times through W, so that the order of the projection is only $x - s$.

The number of tangents to l cutting l once more, is $\gamma = rm + 12r - 14m - 6n$. Of these $2\sigma(\mu - 2\varepsilon - 2)$ lie in w through W. The rest gives the number of circles of curvature touching C once more.

The number of triple points of K is:

$$t = \tfrac{1}{6}\{r^3 - 3r(r + n + 3m) - 58r + 42n + 78m\}.$$

Of these however

$$4\,\frac{(\mu - 2\varepsilon)(\mu - 2\varepsilon - 1)(\mu - 2\varepsilon - 2)}{1.\qquad 2.\qquad 3} - 2\sigma(\mu - 2\varepsilon - 2)$$

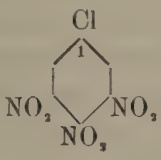
lie in W. The rest gives the number of circles touching C thrice.

If we work out these formulas they get the same form as those of Prof. DE VRIES as is to be expected.

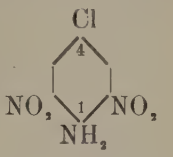
Chemistry. — "*Monochloro-trinitrobenzenes.*" By Prof. A. F. HOLLEMAN.

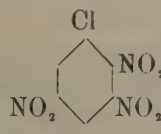
(Communicated at the meeting of June 24, 1922.)

So far only two of the six possible isomers were known, viz. picrylchloride and a product obtained by NIETZKI (see below). For an inquiry into the replaceability of substituents it was required to prepare also the other four isomers. I have only been able so far to lay hands on three, and without doubt I should have waited with the publication of my results till the whole investigation had been completed, if I had not happened to hear that also others are engaged in a study of the same subject.

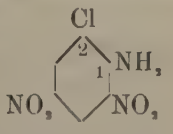
I. 

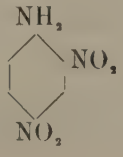
1-chloro-3, 4, 5-trinitrobenzene. This compound is easily accessible; it is indeed surprising that it has not been known long since. The starting-point for its preparation is chlorodinitraniline 1, 4, 2, 6, in which NO_2 was substituted for the NH_2 group according to the method of KÖRNER and CONTARDI. The yield of raw compound amounts to 70% of the theory, and there is only little loss in the purification. The substance may be recrystallized from benzene. It then melts at 168°. Large yellow crystals.

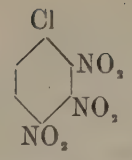


II. 

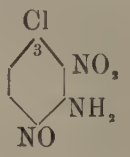
1-chloro-2, 3, 5-trinitrobenzene. This compound is formed on very energetic nitration of 1-chloro-2,3-dinitrobenzene with a mixture of fuming nitric acid and oleum of 50%. The heating of 160—170° is continued for 5 hours. When the mixture is poured out into water, an oil is obtained, in which crystals are formed after some time. By centrifugation these are separated and then recrystallized from alcohol. Melting-point 106°. The structure of this compound was verified by a treatment with alcoholic ammonia, through which 2-chloro-4,6-dinitraniline is obtained, melting-point 159°. This compound is known. Much more easily, however, than according to the methods used up to now it could be prepared by chlorination of 2-4-dinitraniline with $KClO_3$ in hydrochloric acid solution. The entrance of a NO_2-group at the place 5 in 1-monochloro-2,3-dinitrobenzene is very surprising, as this group takes a position at *m* with regard to Cl and at *p* with regard to a nitro-group.

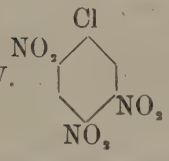




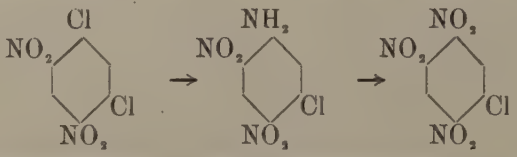
III. 

1-chloro-2,3,4-trinitrobenzene. In the nitration of 1-chloro-2,3-dinitrobenzene by the method given under II, the oil from which II was crystallized contains this third isomer. When the oil stands for a long time, the isomer crystallizes out of it in colourless needles of the melting-point of 69°. They are purified by recrystallization from alcohol. The structure of this compound can also be determined by treatment with alcoholic ammonia. If the action of the ammonia is allowed to last only for a short time, only one of the nitro-groups is replaced by NH_2. The aniline formed is 3-chloro-2, 6-dinitraniline for 1-chloro-2, 4-dinitrobenzene is obtained from it by deamidation. This aniline has the melting-point 112°; it was unknown up to now.



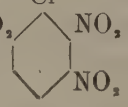
IV. 

1-chloro-3, 4, 6-trinitrobenzene. This compound was already prepared by NIETZKI by nitration of 1-chloro-3,4-dinitrobenzene. On repetition of his experiments it appeared to me that the yield was small, and especially very uncertain, because either the nitration remains incomplete, or the reaction is so violent that total destruction ensues. It is therefore, better to proceed as follows:



The substitution of NH_2 for Cl takes place in alcoholic solution on the waterbath with addition of gaseous ammonia, till a test-sample shows the correct melting-point of 174°. According to KÖRNER and CONTARDI NO_2 can then be substituted for NH_2. The crude product is coloured black. It can be purified by boiling with nitric acid 1.4, followed by recrystallisation from alcohol. The melting-point is 116°, as has been given by NIETZKI.

No method of preparation has as yet been found for the last isomer, the 1-chloro-2,3,6-trinitro-benzene; probably it is also present in the oil obtained in the nitration of 1-chloro-2,3-dinitrobenzene.



Amsterdam, June 1922. *Org. Chem. Lab. of the University.*

Physiology. — "*On Respiratory Oscillations in the Galvanogram of Man.*" By A. A. WEINBERG. (Communicated by Prof. E. D. WIERSMA.)

(Communicated at the meeting of June 24, 1922).

An inquiry into the psycho-physiological significance of the psycho-galvanic reflex, which will ere long be reported in detail, gave rise to the question whether the respiratory arhythmiae in the plethysmogram which result from a predominating influence of the sympathetic nerve, or of the vagus, on the heart [1]), may be attended with oscillations in the so-called rest-current of the galvanogram. In order to set this question at rest the following experimental arrangement was set up.

Our subjects were healthy individuals from 20 to 40 years of age, without any anomalies of the heart or the urine. The current was lead off by non-polarizable electrodes from the baths of a four-cell bath, and was registered with the quick sensitive electrocardiograph of SIEMENS and HALSKE. The non-polarizable electrodes consisted of porous pots filled with a saturated zinc-sulphate solution, with a zinc rod. These pots were placed in the baths, which contained a physiological NaCl-solution heated to body-temperature. The current was recorded by the compensation-method, as the condensator-method [1]) does not enable us to observe the slow oscillations of the current. The sensitivity of the galvanometer, which was controlled for each separate registration, amounted to 4 m.V. per cm. For convenience' sake I selected the three leads which are generally used for taking an electrocardiogram. The method of EINTHOVEN and ROOS [2]), which implies the use of fingerelectrodes and has the advantage of not being complicated by the electrocardiogram, did not yield satisfactory results in these experiments. For further particulars of the procedure of the experiments I refer the reader to my article in the "Nederlandsche Tijdschrift voor Geneeskunde" (l.c.).

With all the subjects thus far examined in this way (fifteen) respiratory oscillations were noticeable in the level of the electric curve. The only requisite was that the subjects had to be in a condition of perfect quiescence, and that their attention be not diverted by anything. Directly when they were more or less pre-

[1]) A. A. WEINBERG, Ned. Tijdschrift v. Geneeskunde; 66, II, 343, 1922.
[2]) W. EINTHOVEN and J. ROOS, Pflüger's Archiv; 189, 126, 1921.

occupied, either in consequence of the experiment or through the after-effect of emotional occurrences, the respiratory oscillations disappeared from the galvanogram, while in the case of still intenser preoccupation other modifications in the level of the curve appeared, which were independent of respiration. All these modifications in the shape of the galvanic curve run parallel with the oscillations in the level of the plethysmogram, either in the same or in the opposite direction. Curve I is an illustration of the respiratory oscillations in the galvanogram.

The following objections may be raised to the hypothesis that these oscillations are connected with the respiratory oscillations in the equilibrium between the sympathetic and the parasympathetic (vagus resp.) nervous system :

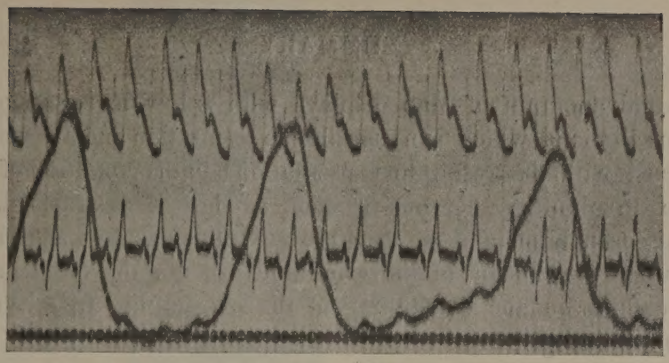
a. The oscillations are effected by the movements of the respiration-muscles.

b. They are caused by the changes in the electrical resistance which are brought about by rhythmic movements of the arm during respiration.

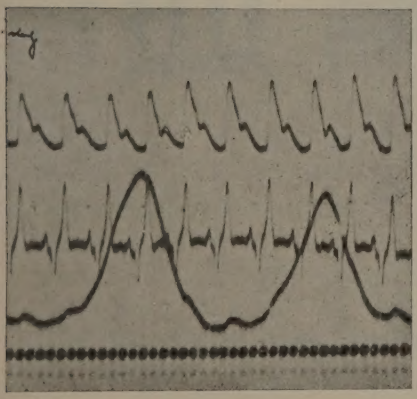
c. They are caused by the respiratory oscillations in the bloodfilling of the extremities.

The first objection is done away with by the fact that in the case of preoccupation the fluctuations disappear (curve II) whereas the movements of the respiration-muscles continue. This phenomenon might likewise tell against the second objection, just as the fact that the subject always rested his hand on the bottom of the arm-baths. Hereby the movements of the upper-arm, which were already none too vigorous at first, were considerably relaxed if not checked entirely. However, with six subjects I have registered the movements of the upper-arm with the aid of a very sensitive tambour affixed to the arm-bath, its rubber membrane, which is provided with a knob, resting on the m. biceps. Hereby it was proved that the movements of the upper-arm do not affect the shape of the galvanogram. This is illustrated in Curve III where the movements of the upper-arm are reproduced graphically ; of oscillations in the level of the galvanic curve, on the other hand, no trace is to be seen. Curve IV further shows that even considerably stronger involuntary arm-movements do not alter the shape of the galvanogram.

Finally, that the electric modifications in respiration cannot be ascribed to the bloodfilling of the extremities is demonstrated by curve V, which exhibits distinct respiratory oscillations of the galvanogram, although the extremities, from which current were derived, had been made bloodless by bandaging.

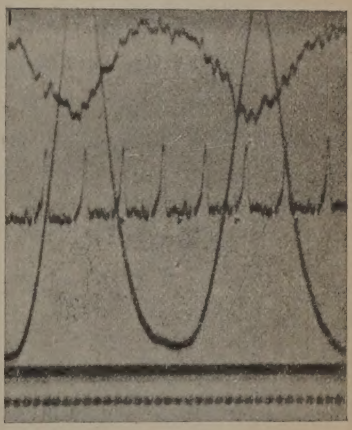


I. G. T. H. ♀ 22 years 3 VI '22. Lead I. Comp. Rest-curve.



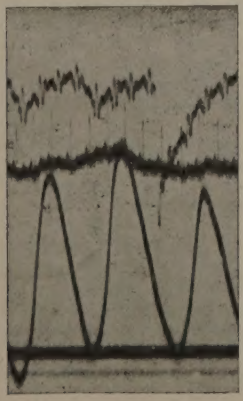
II. The same. 3 VI '22 Lead I. Comp. Pre-occupation curve.

Movement-curve; right upper-arm.

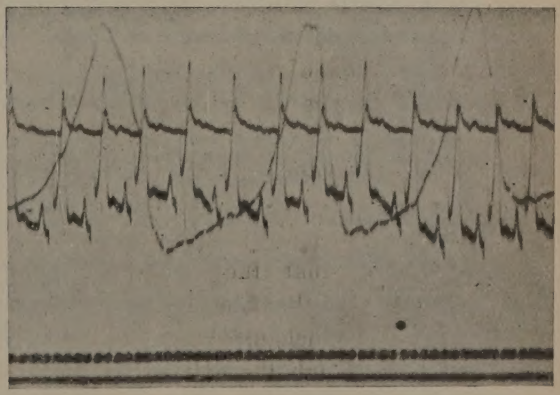


III. S. v. d. G. ♀ 31 years 19 VI '22. Lead II. Comp. Pre-occupation-curve, with control.

Movement-curve; left upper-arm



IV. G. T. ♀ 39 years 6 VI' 22. Lead III. Comp. Rest-curve, with control.



V. D. T. ♂ 21 years 9 VI '22. Lead II. Comp. Rest-curve with dehematized right arm and left leg. Plethysmogram left hand.

SUMMARY.

The galvanogram of Man displays with a low degree of consciousness, oscillations which run parallel to respiration and are very likely connected with the respiratory oscillations in the condition of balance in the involuntary nervous system, as these oscillations disappear with preoccupation and as they are not influenced by the involuntary respiratory movements of the arms and are not the outcome of the modifications in the bloodfilling of the extremities, from which the current is derived.

From the Laboratory for Psychiatry
of the Groningen University.

June 1922.